from

**INDEPENDENT AID**
**NEW YORK CITY**
through

12 JUN 19

.

DISEASES *of* POULTRY

# DISEASES *of* POULTRY

☆

*EDITED BY*

## H. E. BIESTER
*Professor of Veterinary Research and Assistant Director,*
*Veterinary Research Institute, Iowa State College*

AND

## L. H. SCHWARTE
*Professor of Veterinary Research, Veterinary Research Institute,*
*Iowa State College*

### CONTRIBUTORS

| | | |
|---|---|---|
| J. R. BEACH | H. L. FOUST | PETER K. OLITSKY |
| E. R. BECKER | L. T. GILTNER | CARL OLSON, JR. |
| E. A. BENBROOK | E. A. HEWITT | EMMETT W. PRICE |
| K. L. BULLIS | W. R. HINSHAW | L. H. SCHWARTE |
| L. D. BUSHNELL | ERWIN JUNGHERR | M. L. SCOTT |
| JAMES H. BYWATERS | NORMAN D. LEVINE | R. M. SHERWOOD |
| J. R. COUCH | H. C. McDOUGLE | H. J. STAFSETH |
| CHAS. H. CUNNINGHAM | K. F. MEYER | E. L. STUBBS |
| A. J. DURANT | CHAS. MURRAY | HENRY VAN ROEKEL |
| WILLIAM H. FELDMAN | J. L. NOORDSY | NELSON F. WATERS |
| R. FENSTERMACHER | L. C. NORRIS | EVERETT E. WEHR |

☆

*Second Edition, 1948*

ISCP

## THE IOWA STATE COLLEGE PRESS
AMES, IOWA

# FOREWORD

By John R. Mohler, *Chief, Bureau of Animal Industry*
*United States Department of Agriculture*
*Washington, D. C.*

Owners of profitable poultry flocks owe much of their success to diligence in combating diseases. Poultry are subject to a wide variety of infections, some of which respond readily to treatment, whereas others resist control measures and cause discouraging losses.

A knowledge of the characteristics of each disease is necessary, therefore, as the first step in building up an effective barrier against it. Scientific research already has provided much information of this nature, highly useful as a guide throughout the poultry enterprise. Large expenditures of money, time, and labor are still being made by poorly informed, misguided poultrymen in attempting to control disease by futile means.

With the gradual unfolding of scientific knowledge concerning poultry diseases and parasites, the influence of various infections in connection with breeding, nutrition, housing, and management becomes more and more apparent. This volume contains discussions relating to these and allied phases of poultry raising. The authors, who are specialists in their respective fields, encourage a wider application of sound practices recommended for conquering diseases.

This unusually comprehensive book is intended for students, veterinarians, pathologists, and workers in specialized fields. Trained persons usually are best able to make proper diagnoses of disease conditions and to direct proper courses of treatment.

Finally, the suppression of poultry maladies has a public as well as private aspect. The effect of a loss caused by disease is seen and felt first by the flock owner, but the presence of a transmissible infection is a danger to neighboring flocks as well. Morover, large numbers of small individual losses add up to a substantial national total. I trust, therefore, that this book may have a far-reaching influence in improving poultry health, with benefits to the poultry industry and the national welfare.

<div align="right">JOHN R. MOHLER</div>

*Washington, D. C.*
*April, 1943*

# PREFACE TO SECOND EDITION

The recent advances in diseases of poultry and related fields of biology necessitated revision and enlargement of the first edition of *Diseases of Poultry*. Essentially the same policy that was adopted in the preparation of the first edition was followed in the revision.

The editors are deeply indebted to the collaborators for their continued and unfailing support of the undertaking. Their loyalty and interest constitute the chief motivating forces in maintaining the continuity of the project.

The preparation of the second edition would not have been possible without the encouragement of Dean H. D. Bergman and President Charles E. Friley.

The patience, helpfulness, and deep interest shown in the preparation of the second edition by Mr. H. E. Ingle and other members of the Iowa State College Press staff made our relations with the Press a pleasant experience.

The splendid cooperation of Mrs. Mildred E. Hicks, Mrs. L. M. Smith, Director R. W. Orr, and other members of the Library staff aided greatly in the completion of the second edition. The outstanding collection in comparative medicine and allied fields of the Iowa State College Library greatly facilitated the diversified bibliographic work.

The intense interest and cooperation of Mrs. Barbara Ives in all phases of the work proved invaluable.

H. E. BIESTER
L. H. SCHWARTE

*Ames, Iowa*
*June, 1948*

# TABLE OF CONTENTS

[ix]

# THE AUTHORS

BEACH, J. R., Department of Veterinary Science, University of California, Berkeley. Chap. 13 (p. 345) —*Infectious Coryza.* Chap. 19 (p. 475) —*Infectious Bronchitis.* Chap. 20 (p. 481) —*Infectious Laryngotracheitis.* Chap. 21 (p. 489) —*Avian Pneumoencephalitis (Newcastle Disease).*

BECKER, E. R., Department of Zoology, Iowa State College, Ames. Chap. 35 (p. 863) —*Protozoa.*

BENBROOK, E. A., Department of Veterinary Pathology, Iowa State College, Ames. Chap. 31 (p. 715) —*External Parasites of Poultry.*

BULLIS, K. L., Department of Veterinary Science, University of Massachusetts, Amherst. Chap. 17 (p. 403) —*Diseases Caused by Fungi.*

BUSHNELL, L. D., Department of Bacteriology, Kansas State College, Manhattan. Chap. 10 (p. 277) —*Fowl Typhoid.*

BYWATERS, JAMES H., Virginia Agricultural Experiment Station, Blacksburg. Co-author Chap. 3 (p. 43) —*Poultry Genetics as Related to Pathology.*

COUCH, J. R., Department of Poultry Husbandry; Texas Agricultural Experiment Station, College Station. Co-author Chap. 7 (p. 157) —*Vitamins and Vitamin Deficiencies.*

CUNNINGHAM, CHAS. H., Department of Bacteriology and Public Health, Michigan State College, East Lansing. Chap. 25 (p. 567) —*Fowl Pox.*

DURANT, A. J., Department of Veterinary Science, University of Missouri, Columbia. Co-author Chap. 36 (p. 947) —*Diseases of the Digestive System.*

FELDMAN, WILLIAM H., Division of Experimental Medicine, Mayo Foundation, Rochester, Minnesota. Chap. 12 (p. 311) —*Tuberculosis.* Co-author Chap. 30 (p. 637) —*Neoplastic Diseases of the Chicken.*

FENSTERMACHER, R., Diagnosis Laboratory, University Farm, St. Paul, Minnesota. Chap. 9 (p. 247) —*Paratyphoid Infections.*

FOUST, H. L., Department of Veterinary Anatomy, Iowa State College, Ames. Chap. 1 (p. 1) —*Anatomy.*

GILTNER, L. T., Pathological Division, Bureau of Animal Industry, United States Department of Agriculture, Washington, D. C. Chap. 24 (p. 561) — *Equine Encephalomyelitis Virus in Birds.*

HEWITT, E. A., Department of Veterinary Physiology and Pharmacology, Iowa State College, Ames. Chap. 2 (p. 29) —*Digestion.*

HINSHAW, W. R., Department of Veterinary Science, University of California, Davis. Chap. 5 (p. 89) —*Principles of Disease Prevention.* Chap. 40 (p. 1015) —*Diseases of the Turkey.*

JUNGHERR, ERWIN, Department of Animal Diseases, University of Connecticut, Storrs. Chap. 18 (p. 421) —*The Avian Leukosis Complex.* Chap. 29 (p. 623) —*Avian Monocytosis.*

LEVINE, NORMAN D., Department of Veterinary Pathology and Hygiene, College of Veterinary Medicine, University of Illinois, Urbana. Chap. 15 (p. 369) —*Listerellosis, Botulism, Erysipelothrix, and Goose Influenza.*

McDOUGLE, H. C., Department of Veterinary Science, University of Missouri, Columbia. Co-author Chap. 36 (p. 947) —*Diseases of the Digestive System.*

MEYER, K. F., The George Williams Hooper Foundation for Medical Research, University of California, San Francisco. Chap. 22 (p. 513) —*Psittacosis (Ornithosis).*

MURRAY, CHAS., Division of Veterinary Medicine, Iowa State College, Ames. Chap. 11 (p. 299) —*Fowl Cholera.*

NOORDSY, J. L., Veterinary Research Institute, Iowa State College, Ames. Chap. 16 (p. 389) —*Streptococcosis, Staphylococcosis, Arthritis, Colibacillosis, and Hjärre's Disease.*

NORRIS, L. C., Department of Poultry Husbandry and School of Nutrition, Cornell University, Ithaca, N. Y. Co-author Chap. 6 (p. 115) —*Proteins, Carbohydrates, Fats, Fiber, Minerals, and Water in Poultry Feeding.*

OLITSKY, PETER K., The Laboratories of the Rockefeller Institute for Medical Research, New York City. Chap. 23 (p. 551) —*Avian Encephalomyelitis (Epidemic Tremor).* Chap. 27 (p. 615) —*Foot-and-Mouth Disease in Fowl.*

OLSON, CARL, JR., Department of Animal Pathology and Hygiene, University of Nebraska, Lincoln. Chap. 4 (p. 69) —*Avian Hematology.* Co-author Chap. 30 (p. 637) —*Neoplastic Diseases of the Chicken.*

PRICE, EMMETT W., Zoological Division, Bureau of Animal Industry, United States Department of Agriculture, Washington, D. C. Chap. 34 (p. 839) — *Trematodes of Poultry.*

SCHWARTE, L. H., Veterinary Research Institute, Iowa State College, Ames. Chap. 28 (p. 619) —*Rabies and Infectious Equine Anemia.* Chap. 37 (p. 961) —*Poultry Surgery.* Chap. 38 (p. 975) —*Vicious Habits and Miscellaneous Conditions.* Chap. 39 (p. 987) —*Poisons and Toxins.*

SCOTT, M. L., Department of Poultry Husbandry and School of Nutrition, Cornell University, Ithaca, New York. Co-author Chap. 6 (p. 115) — *Proteins, Carbohydrates, Fats, Fiber, Minerals, and Water in Poultry Feeding.*

SHERWOOD, R. M., Department of Poultry Husbandry, Texas Agricultural Experiment Station, College Station. Co-author Chap. 7 (p. 157) —*Vitamins and Vitamin Deficiencies.*

STAFSETH, H. J., Department of Bacteriology and Hygiene, Michigan State College, East Lansing. Chap. 14 (p. 353) —*Brucellosis, Anthrax, Pseudotuberculosis, Tetanus, and Vibrio Infections.*

STUBBS, E. L., Department of Pathology, School of Veterinary Medicine, University of Pennsylvania, Philadelphia. Chap. 26 (p. 603) —*Fowl Pest.*

VAN ROEKEL, HENRY, Department of Veterinary Science, Massachusetts Agricultural Experiment Station, Amherst. Chap. 8 (p. 203) —*Pullorum Disease.*

WATERS, NELSON F., United States Regional Poultry Research Laboratory, East Lansing, Michigan. Co-author Chap. 3 (p. 43) —*Poultry Genetics as Related to Pathology.*

WEHR, EVERETT E., Zoological Division, Bureau of Animal Industry, United States Department of Agriculture, Washington, D. C. Chap. 32 (p. 759) — *Nematodes and Acanthocephalids of Poultry.* Chap. 33 (p. 809) —*Cestodes of Poultry.*

# CHAPTER ONE

# ANATOMY

By H. L. Foust, *Department of Veterinary Anatomy,*
*Iowa State College, Ames, Iowa*

☆ ☆ ☆

## EMBRYOLOGY

The scope of this chapter does not permit discussion of the embryology of the fowl. Amongst those who have published in this field appear the following names: His, 1868; Duval, 1889; Keibel, 1900; Foster *et al.*, 1902; Minot, 1903; Lillie, 1918; Kerr, 1919; Bailey and Miller, 1929; Needham, 1931; McEwen, 1931; Champy, 1934; Bradley, 1938; Huetner, 1941; Venzke, 1942; Patten, 1946; Arey, 1946.

## THE SKELETON

The bones of fowls, being especially rich in calcium salts, are very dense. In many the marrow is displaced by air spaces which are in communication with the respiratory tract. According to Vermeulen (1929) there are no diaphyseal-epiphyseal cartilages. Figure 1.1 shows the major skeletal features.

## THE SKULL

The bones of the head conform well to its peculiar shape. The bones of the face are combined into the upper beak which is movably connected with the skull. The two orbits are very large, being separated from each other by the perpendicular plate of the ethmoid. The palatine processes of the maxillae do not fuse; consequently, the palate is cleft and the floor of the nasal cavity incomplete. Great mobility of the occipito-atlantal joint is facilitated by the unpaired condyle of the occipital bone.

## THE VERTEBRAE

The pigeon has twelve, the chicken thirteen or fourteen, the duck fourteen or fifteen, and the goose seventeen or eighteen cervical vertebrae. The large size of the articular processes makes for the great motility of this region. In the thoracic region the vertebral column is relatively short. It is composed of seven vertebrae in pigeons and chickens, and of nine in ducks and geese. Of these in chickens the second to the fifth are fused, and the first and sixth are free. The seventh is fused with the first lumbar. In the fused group

the spinous and transverse processes form nearly complete plates. The lumbar and sacral vertebrae are fused, being composed of some 14 segments. The last thoracic and the first coccygeal segments are joined to each end of the mass. Ridges on the ventral surface represent the transverse processes of the segments. The coccygeal vertebrae are in number some five or six in the
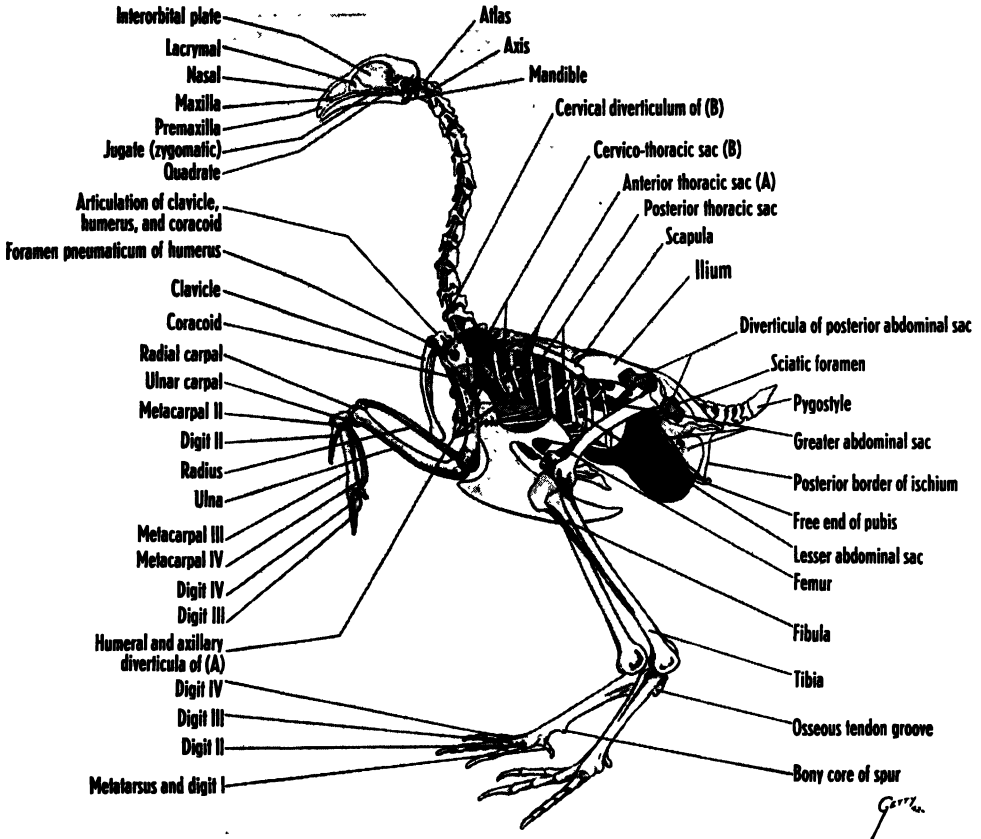


FIG. 1.1. Skeleton of chicken with semischematic outlines of the air sacs. One leader line for each sac indicates its bronchial connection with the lung.

chicken and seven to eight in the pigeon, duck, and goose. There is a fusion of several vertebrae to form the last segment of the coccygeal group. This segment is called the pygostyle. This group is capable of considerable movement and forms the skeletal support for the tail feathers.

## THE RIBS AND STERNUM

There are seven pairs of ribs, of which the first 2 and the last 1 are asternal. The sternal segments of the sternal ribs are bony and are united to the vertebral portions by joints. On the posterior borders of the second to

the sixth vertebral segments of the ribs, are flat processes (Processus uncinati) which may overlap the segments next following.

The sternum is long and broad. Since it extends posteriorly to the pelvic region it, by supporting viscera, compensates somewhat for deficiencies in the abdominal muscles. Lateral to the crest on each side, the posterior end presents a more or less complete foramen in the pigeon and goose. In the duck and chicken lateral incisures are present; in the chicken these are double since the lateral processes are bifid. Size of the sternal crest seems to parallel flying ability. At the anterior end lateral to the rostrum are facets for articulation with the proximal extremities of the coracoid bones. The dorsal surface presents several foramina which connect air spaces in this bone with the respiratory organs.

The observations of Cuvier (1832) on the ossification of the avian sternum are of interest. In the chicken at the seventeenth day of incubation, the postero-lateral processes had begun to ossify. At 19 days a center had appeared in the anterior end of the keel near its base. At hatching, the postero-lateral processes had ossified in the greater part of their length, and the antero-lateral angles had become osseous.

At 30 days of age the costal branches of the postero-lateral processes had nearly ossified, while the median branch remained cartilaginous for a long time. At 140 days the rostrum had become ossified by an extension of the process from the body of the bone (growth as apophysis and not epiphysis). At 72 days the centers of the costal processes and the postero-lateral processes had not yet fused with each other or with the body of the bone, but at 93 days the centers of the same side had fused. The five growth areas, one for the body and one each for the costal and postero-lateral processes were nearly all fused into one bone by 113 days, and at five or six months even the posterior end of the metasternum was found to be completely ossified. This method of ossification of the sternum was said to occur in gallinaceous birds, while in others, such as the swan, goose, duck, pigeon, and birds of prey, the process was a bilateral one, the first evidence of ossification having appeared in the antero-lateral angles of the sternum. From these two points the process extended medially and posteriorly until complete. The process began in the duck rather late, 40 days after hatching. At 115 days the oval foramina were nearly encircled. The encirclement was completed only in older individuals at which time the sternum was said to be completely ossified (Cuvier, 1832).

### SKELETON OF THE WING

Scapula, coracoid, and clavicle compose the shoulder girdle. The scapula, long, narrow and slightly curved, lies dorsal to the ribs. It articulates with the coracoid at an acute angle. The coracoid is the largest bone in this group and articulates by its proximal end with a facet on the sternum. This proxi-

mal end presents a foramen which connects with the anterior thoracic air sac. The distal end of this bone has a hooklike process which together with the proximal ends of the humerus and scapula forms a ring of bone (Foramen triosseum) through which passes the tendon of the supracoracoid muscle which is an elevator of the wing. Anterior to the coracoid is the clavicle which by its proximal end articulates with the coracoid and humerus and by its distal end meets its fellow to form a forked bone (the furculum, wish bone). A ligament unites the furculum to the rostrum of the sternum. This ligament was said by Vermeulen (1929) to ossify in some birds.

The humerus presents on the medial side of the proximal end, a Foramen pneumaticum (Fig. 1.1) which connects with the anterior thoracic air sac. The ulna is much larger than the radius.

In the adult a radial and an ulnar element compose the bones of the carpus. Following the method commonly used for description of reduced members of metacarpal bones and digits in mammals, there would be in the chicken the second, third, and fourth incompletely developed metacarpal bones. The second is very short and is attached to the fused proximal ends of the third and fourth. The latter two are also fused at their distal ends. Of the digits corresponding to the metacarpal bones, the second and third are each composed of two phalanges, and the fourth has one phalanx.

### SKELETON OF THE LEG

Ilium, ischium, and pubis compose the very incomplete pelvic girdle. The ilium and ischium are extensively fused with the regional vertebrae, and in the concavities of the ventral surfaces of these two bones the kidneys are lodged. A large sciatic foramen perforates the ischium posterior to the acetabulum. The pubis is a long, slender bone beginning antero-ventral to the acetabulum by a knob and extending posteriorly with its free end projecting beyond the ischium. Slightly postero-ventral to the acetabulum, it is separated by the obturator foramen from the ischium. There is attachment between pubis and ischium for a short distance posterior to the foramen, beyond which the shaft and extremity of the pubis are free.

The femur, patella, fibula, and tibia very closely resemble those of mammals. Since the tarsal bones fuse early with the tibia and metatarsal bones, there are no separate tarsal bones. The tarsal joint is a ginglymus or hinge joint composed in the adult of the fused bones on the distal end of the tibia and those on the proximal end of the metatarsus.

The metatarsus is composed of a much-shortened first metatarsal bone connected by a ligament to the medio-plantar border of the fused second, third, and fourth metatarsal bones. Very close and proximal to the first, there is in the male on the medial side of the shaft of the fused mass a hooked

process which is the base of the spur. The distal end of the metatarsus has three condyles which articulate with the first phalanges of the second, third, and fourth digits.

In the chicken the first digit of the foot is directed posteriorly and carries two phalanges. The second, third, and fourth digits are directed anteriorly and have, respectively, three, four, and five phalanges. In the parrot the first and fourth digits are directed posteriorly, while in swimming birds all four digits are anterior. Chamberlain (1943) has published an excellent atlas on the osteology, arthrology, and myology of the fowl.

### MUSCULAR SYSTEM OF FOWLS

There are some groups of skeletal muscles in fowls which, because they have certain peculiarities, will be mentioned.

Cutaneous muscles have a somewhat wider distribution in fowls than in mammals. The patagial muscle which extends into the skin fold (patagium) of the wing, and the cutaneous muscles supplying the skin which supports the tail feathers may be mentioned especially.

The pectoral muscles are especially highly developed in birds which do much flying. There is a superficial one which depresses the wing and a deeper one called by Bradley (1938) the supracoracoid muscle. This latter one has a tendon of insertion which passes through the Foramen triosseum at the shoulder joint to be inserted on the humerus near its head. This muscle is an elevator of the wing.

The abdominal muscles are very thin sheets of muscle and aponeurosis. The obliquus abdominis externus muscle takes origin from the uncinate processes and from the lateral surfaces of the ribs at the level of the processes. The attachment begins at the second rib and extends posteriorly along the entire lateral border of the Os coxae. The insertion is to the linea alba, the metasternum, the branches of the postero-lateral processes of the sternum, and the major pectoral muscle. The fleshy portion, about 1½ inches in width, extends from a line connecting the posterior ends of the metasternum and pubic bones to its attachment on the second rib. Its ventral border blends with the pectoralis major muscle.

The rectus abdominis muscle lies deeply to the posterior portion of the fleshy part of the external oblique muscle. Its origin is from the pubis and from the aponeurotic sheet common to the muscle and its fellow of the opposite side and to the external oblique and transverse abdominal muscles. It inserts on the metasternum and on the postero-lateral process of the sternum. Its fleshy part begins near its origin from the pubis and forms a thin sheet which extends to the posterior extremities of the metasternum and of the posterior lateral process of the sternum.

The obliquus abdominis internus muscle is a thin fleshy sheet taking its origin from the ventral border of the Os coxae anterior to the rectus. It inserts on the posterior border of the last rib. It forms a triangular sheet which fills the angle between the last rib and the ventral border of the tuber coxae.

The transverse abdominis muscle arises from the aponeurosis common to it and its fellow and from the ventral border of the Os coxae and from the proximal portion of the deep surface of the last two ribs. It inserts to the linea alba, and its aponeurosis fuses with that of the rectus.

These four muscles form the lateral wall of the abdominal cavity. The combined aponeuroses of the external oblique, the rectus, and the transverse muscles form a triangular aponeurotic sheet with its apex at the posterior end of the metasternum and its base between the posterior ends of the pubic bones. This sheet thus forms the floor of the abdomen.

Ossification of many of the tendons of the muscles of the hind limb is common in many species of fowl.

The diaphragm, according to McLeod and Wagers (1939), is composed of two parts: (1) A thoraco-abdominal tendinous sheet which is attached to the sternum, to the sixth and seventh ribs, and to the sixth thoracic vertebra. It separates the thoracic and abdominal cavities and has two paired openings for the abdominal air sacs. (2) The pulmonary part, according to the same two authors, divides the thoracic cavity into dorsal and ventral portions. It has a muscular portion (Mm. costo-pulmonales) attached to the free ends of the first and second pairs of ribs, to the posterior border of the sixth rib, and to the upper parts of the sternal ends of the intervening ribs. It is attached medially to the ventral spines of the thoracic vertebrae. Its foramina afford passage-ways for air sacs, and for the bronchi, and for the pulmonary vessels and nerves. Its posterior portion is fused with the dorsal part of the thoraco-abdominal part of the diaphragm. Our dissections have corroborated the description of McLeod and Wagers (1939). Somewhat similarly, Chauveau and Arloing (1891) have referred to the two-part avian diaphragm.

## THE DIGESTIVE SYSTEM
### THE BEAK

The discussion of the microscopic structure of the parts of the digestive system will follow Calhoun's (1933) descriptions. The beak, both upper and lower segments, is composed of a superficial epidermis. This has the four typical strata ordinarily ascribed to it. Derma or corium intervenes between epidermis and periosteum. This layer contains blood vessels and nerves, and some touch corpuscles. The bone of the upper beak is the Os incisivum (praemaxillae), that of the lower, the Os dentale (corpus mandibulae).

The lining membrane is similar to that of mammals. Many posteriorly directed papillae are present in the mucosa of the hard palate, which presents a median cleft. Lymphoid tissue is present in the lamina propria of older birds.

### THE TONGUE

The dorsum of the tongue has a mucous membrane which, although uneven, is devoid of papillae except posteriorly where there is a transverse row. The stratified squamous epithelium on the dorsum is highly kerati-



FIG. 1.2. Section of mid-portion of hard palate of chicken. (Calhoun.)

1—Epithelium.  4—Fat.
2—Tunica propria.  5—Central cavity of lobes of medial palatine salivary gland.
3—Submucosa.  6—Papilla.

nized. It continues over the tip to the ventral side where soon it becomes less cornified and smooth. Lymphoid tissue may be present in this part of the tongue of older birds. The muscles are poorly developed and are arranged about the arrow-shaped end of the entoglossal bone which is the anterior end of the hyoid bone. This entoglossal bone is continued posteriorly by the basihyoid, a cylindrical centrally located segment.

### THE SALIVARY GLANDS

The salivary glands all have a similar structure. They may be classified as branched tubular glands (Fig. 1.2). The tubules of each lobule open into a central cavity which is continuous with an excretory duct.

Calhoun (1933) following Schauder's (1923) descriptions, located the following glands:

"(a) Glands at the bottom of the oral cavity.
    1. Anterior submaxillary: largely developed, paired glands in the angle between the rami of the lower jaw.

   2. Posterior submaxillary in group of 3:
      a. antero-lateral, lying medial to the Os dentale;
      b. intermediary, caudo-ventral to a;
      c. postero-medial, postero-medial to and connecting with the intermediary
         group.
  (b) Glands of angle of mouth.
   3. Angularis oris gland (Cholodkowsky, 1892) ; lying in the angle of the beak, a
      small, three-cornered gland area.
  (c) Glands of the tongue.
   4. Anterior lingual: at the side of, in the middle of, and in the posterior part of
      the tongue.
   5. Posterior lingual: on the dorsal surface of the base of the tongue.
  (d) Glands of the roof of the mouth.
   6. Paired glands joining medially in the hard palate lying anterior to the
      posterior nares. (Maxillary of Heidrich, 1905.)
   7. Medial and lateral palatine glands: extending longitudinally to the posterior
      nares.
   8. Sphenopterygoid: in the roof of the pharynx.
  (e) Glands of the pharyngeal canal.
   9. Cricoarytaenoideae: lying lateral to the larynx in the submucosa of the
      cutaneous mucous membrane."

Although Calhoun (1933) found only mucous cells in these glands, Vermeulen (1929) reported that in some granivorous birds, serous cells had been found in some glands. Leasure and Link (1940) said they found amylase in the saliva of the hen. Lymphoid tissue may be found both inter- and intralobular in most of the salivary glands of adult birds.

### THE PHARYNX

The posterior-most transverse row of palatine papillae and the row on the base of the tongue may be taken as a division line between mouth and pharynx. The mucous membrane is similar in structure to that of the mouth. Dorsally on the median plane is a slit which provides a common opening for the pharyngeal ends of the eustachian tubes. Continuity with the nasal cavity is through the cleft of the hard palate.

### THE ESOPHAGUS

The structure of the esophageal wall is similar to that of mammals. The thick epithelial layer of the mucosa is highly keratinized. Extending through this layer and into the lamina propria are large mucous glands. Some lymphoid tissue may be found in the lamina propria. The muscularis mucosae is very thick. The submucosa is thin. The inner circular lamina of the muscular tunic is quite thick, while the outer longitudinal layer is composed of sparse bundles of smooth muscle. The tunica adventitia is composed of loose fibrous tissue.

### THE CROP

The esophagus of ducks and geese has in its cervical portion a long spindle-shaped dilatation. In pigeons and chickens a ventral diverticulum marks the junction of cervical and thoracic portions. In the chicken this

crop or ingluvies has a structure similar to that of the esophagus with the exception that it is glandless in its greater part, and its greater curvature has a sparse blood supply. The crop of the pigeon has two lateral lobes. The structure of the wall is similar to that of the esophagus. In the female pigeon which has been setting for 8 days, hypertrophy of the wall has already begun. This remains until several days after hatching. The most marked change is in the increased number of cells of the epithelial layer which becomes markedly folded. The superficial cell-layers of the epithelial layer become laden with fat and are desquamated into the lumen to form pigeon "milk" (Beams and Meyer, 1931). A similar process is said to occur in the male during about the same period.

In the portion of the esophagus between the crop and proventriculus, the epithelium decreases in thickness toward the junction with the proventriculus at which point there is an abrupt transition to the simple columnar type of epithelium which lines the gut from this point to the anus. There is a gradual transition from the characteristic mucous glands of the eosphagus to the type found in the proventriculus.

### THE PROVENTRICULUS

The proventriculus, although the glandular stomach of common fowl, differs from the similar mammalian structure, in that its lumen is scarcely larger than that of the esophagus. Its storage capacity is therefore very limited.

There are the four typical layers in its wall. Simple tubular glands lined with mucous cells are found in the superficial part of the mucous membrane. The muscularis mucosae is broken by deeper glands with some peculiar characteristics. These glands are composed of lobules of tubular glands (Fig. 1.3). In the center of each lobule is a cavity which receives the glandular secretion and is continuous with an excretory duct. Several excretory ducts empty into the lumen of the proventriculus through large papillae. The free ends of the cells of the tubular glands are directed toward the central cavity and do not touch adjacent cells, thus giving a serrated appearance to the glandular epithelium. According to Calhoun (1933), cells of the surface epithelium, cells of the epithelium of the collecting canals, and cells of the epithelial lining of the distal third of the excretory ducts of the deeper glands, all are of a mucous character.

There is but a thin layer of submucosa separating the muscularis mucosae from the muscular tunic. This muscular tunic is composed of the two typical layers. The adventitia is a loose connective tissue.

### JUNCTION OF PROVENTRICULUS AND GIZZARD

In this connecting piece the deeper proventricular glands end abruptly, while the superficial glands gradually increase in length and take on the char-

acters of the glands of the gizzard. The propria becomes continuous with the submucosa of the gizzard. Coincidental with this transition, the superficial portion of the muscularis mucosae ceases, and the deeper part fuses with the inner circular lamina of the muscular tunic of the gizzard. The outer longitudinal layer of the muscularis of the proventriculus ceases at the gizzard,
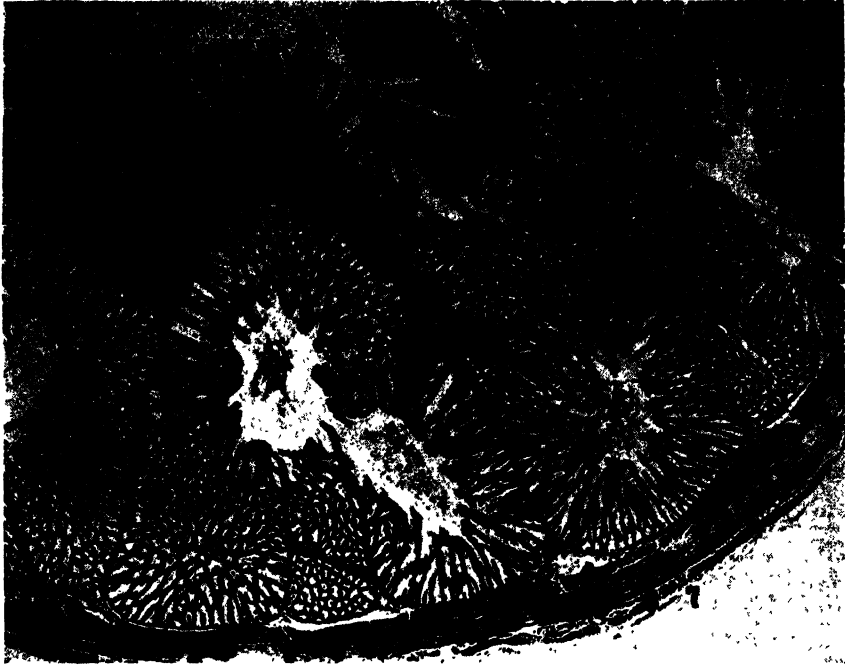


FIG. 1.3. Cross section of proventriculus of chicken. (Calhoun.)

1—Surface tubular glands lined          4—Muscularis mucosae.
   with columnar epithelium.            5—Circular layer of muscular tunic.
2—Tunica propria.                       6—Longitudinal layer of muscular tunic.
3—Gland lobule.                         7—Adventitia.
        Between 4 and 1, a cavity of a gland lobule empties into the lumen.

while the circular layer of the proventriculus becomes continuous with the muscle of the gizzard.

### THE GIZZARD

The gizzard (ventriculus) is a spheroidal organ, flattened in the lateral direction. Its two lateral sides are biconvex discs with aponeurotic vertices. There is a dorsal and a ventral muscular mass attached to the aponeuroses just mentioned. These have been termed Musculi lateralis, dorsalis et ventralis by Schauder (1923). He also describes Musculi intermedii lying on the anterior and posterior culs-de-sac. These muscular masses are of a red

color but nonstriated. There is no muscularis mucosae, thus the propria and submucosa are continuous. There are long, simple tubular glands extending into the propria (Fig. 1.4). These glands are lined with a low cuboidal epithelium. The lumina of the glands contain a substance which, according to Calhoun (1933), responded to a keratohyaline stain. This material with



Fig. 1.4. Section through entire wall of gizzard of chicken. (Calhoun.)
1—Horny layer.  3—Submucosa.  5—Serosa.
2—Glands in tunica propria.  4—Muscular tunic.

cellular debris forms the so-called "horny" layer of the gizzard (Fig. 1.4). It presents wavy lines parallel to the surface and wider lines perpendicular to the surface. These latter appear to coincide in position with the material in the tubular glands. The duodenal and esophageal orifices are quite close together on the antero-dorsal surface of the organ.

In the transitional part of the gizzard leading into the small intestine, Calhoun (1933) found Brunner's type of glands to be present. Schauder (1923) says these glands are pyloric in type.

### THE SMALL INTESTINE

The intestine of the fowl, although similar to that of mammals, differs markedly in some parts. It is about five to six times the length of the body

(Schauder, 1923). Carnivorous birds have a shorter bowel than do granivorous birds.

The duodenum, which does not have glands of Brunner, presents a loop supporting the pancreas and is generally considered to terminate at the entrance of the bile and pancreatic ducts. The jejunum and ileum are supported by a mesentery and bounded by air sacs which separate them from the abdominal wall. There is often a diverticulum on this portion of the intestine which is a remnant of the yolk stalk. The wall of the intestine from outside to inside comprises: (1) serous tunic, (2) muscular tunic (outer longitudinal, inner circular), (3) a very thin submucosa, (4) mucosa. The muscularis mucosae was described by Calhoun (1933) as having an inner longitudinal and an outer circular layer. The propria contains considerable lymphoid tissue and lymph nodules. Crypts of Lieberkühn open into the lumen of the gut between the bases of adjacent villi. Many of the villi branched, according to Calhoun (1933), and the taller ones were found in the duodenal and ileal portions.

At the termination of the ileum there is a fold of mucous membrane into which Calhoun (1933) found a sphincter muscle projecting.

### THE CECA AND LARGE INTESTINE

Beyond the small intestine the bowel presents two retrograde portions (the ceca) and a continuing portion (the large intestine). The paired ceca extend anteriorly for some 9 inches parallel to the ileum, to which they are attached by peritoneal folds. According to Calhoun (1933) the villi were quite well developed near the mouths of the sacs, shorter and broader in the midportions, and in the fundus parts the villi were low and blunt. The glands of Lieberkühn are poorly developed. In older birds lymph nodules and much lymphoid tissue are present in the propria.

### THE RECTUM

Opinions differ in regard to the presence of a colon in fowls. The term, rectum, will here be applied to that portion of the bowel between the cecal orifices and the beginning of the cloaca. The structure of the rectal wall resembles that of the small intestine. The glands of Lieberkühn are, however, much smaller and fewer. A slight constriction usually is present to mark the termination of the rectum.

### THE CLOACA

Usually quite definite circular folds delimit the three portions of the cloaca (the coprodaeum, the urodaeum, and the proctodaeum). The coprodaeum is the passageway between the rectum and urodaeum. The urodaeum continues the passageway between the coprodaeum and the proctodaeum and, in addition, receives the ureters and genital tubes. Between

the urodaeum and the proctodaeum, the limiting fold on the dorsal side guards the entrance to the bursa of Fabricius. The wall of the cloaca has a structure similar to that of the rectum and small intestine. Villi and circular folds are evident, according to Calhoun (1933).

## THE BURSA CLOACAE (OF FABRICIUS)

The Bursa cloacae in the chicken, extending dorsally from the roof of the proctodaeum, attains a size of 2 to 3 × 1½ cm. in four to five months accord-



FIG. 1.5. Section of fold from wall of bursa of Fabricius of chicken of 7 months. (Calhoun.)
1—Trabecula of tunica propria.
2—Lymph nodule.
3—Columnar epithelium.

ing to Schauder (1923), and at about ten months has nearly disappeared. Vermeulen (1929) says the bursa atrophies with the beginning of puberty. The wall of the bursa has a serous tunic and a muscular tunic. Both are typical. The mucosa is much folded, and these folds contain great numbers of lymph nodules (Fig. 1.5). The nodules, Calhoun (1933) found, have a typical lymph-follicle structure (dense periphery with a lighter center). The epithelial layer is composed of modified columnar cells.

## THE ANUS

The structure of the anus of the fowl resembles that of mammals, in that there is the transition from simple columnar epithelium to stratified

squamous epithelium. The muscularis mucosae ceases. A sheet of cross-striated muscle making its appearance at the level of the bursa of Fabricius, extends in the fused propria and submucosa to the borders of the dorsal and ventral anal lips (Calhoun, 1933).

The vascular and nerve supplies of the digestive tube resemble those of mammals.

## THE LIVER

The microscopic structure of the liver varies little, according to Calhoun (1933), from that of mammals. A cystic duct and a hepatic duct both empty into the intestine near the terminations of the pancreatic ducts (Calhoun, 1933).

## THE PANCREAS

The pancreas of the chicken is a compound tubulo-acinar gland with a structure similar to that found in mammals. Calhoun (1933) found islets of Langerhans in the material which she studied. She also found three pancreatic ducts.

## THE RESPIRATORY SYSTEM
### THE NOSTRILS AND NASAL CAVITIES

The anterior naris, elliptical in outline with the long axis antero-posterior, may be guarded by small feathers. The two nasal cavities are separated from each other by a septum composed of bone and cartilage. Three mucous-membrane-covered plates project from the walls; these correspond to mammalian turbinates or nasal conchae. On each lateral wall of the nasal cavities ventral to the middle turbinate and at the level of the anterior end of the palatine cleft, is the opening of the naso-lacrimal canal. Each posterior naris is connected with the cavity of the pharynx through the palatine cleft.

### THE LARYNX AND TRACHEA

The larynx is not guarded by an epiglottis. The larynx of the fowl often called the cranial larynx also is devoid of thyroid cartilages and vocal cords. The cricoid cartilage is segmented.

The cartilaginous rings of the trachea are complete. Outside the tube, Vermeulen (1929) says, there are bundles of skeletal muscles running longitudinally. The mucous membrane contains a pseudostratified columnar epithelium with mucous alveolar glands. Two bronchi continue the air passageway into the lungs. At the point where these begin is the caudal larynx, sometimes called the broncho-tracheal larynx, and syrinx. This is the organ of voice in the bird. A median crest at the bifurcation of the two bronchi surmounted by an elastic lamina on each of its lateral sides together with similar membranes laterally in each bronchial wall serve as vocal cords.

## THE LUNGS

The lungs extend from the first ribs to the anterior poles of the kidneys. The medio-lateral dimension of the lungs is relatively short; consequently, they do not project far into the thoracic cavity. The ventro-medial surface is covered in part by the pulmonary diaphragm, while the dorso-lateral surface is deeply grooved by the ribs. The lungs are not divided into lobes, as in many mammals, but distinct lobules are formed about the terminal loops of the tertiary bronchi.

The bronchi may be considered to be divided into primary, secondary, and tertiary segments. The primary and secondary bronchi each are connected with air sacs. The tertiary bronchi terminate by breaking up into great numbers of anastomosing air tubes or capillaries (as shown in Fig. 2, Taf. II, Fischer, 1905). The epithelial lining of these air capillaries may be considered as respiratory epithelium of the lung proper. Running between the epithelial layers of adjacent air tubes are the capillaries of the pulmonary circulation.

The air sacs which connect with the primary and secondary bronchi have walls of a thin inner mucosa continuous with that of the bronchi and an outer serous layer of either pleura or peritoneum. Several different terms have been applied to the various air sacs. Those proposed by McLeod and Wagers (1939) seem from a topographical viewpoint to be desirable and will be followed here. Figure 1.1 illustrates the locations of the various air sacs.

The paired thoraco-cervical sacs are found at the cervico-thoracic junction. Prolongations extend anteriorly through each Canalis transversarius as far sometimes as the third cervical vertebra, while those going posteriorly may reach the fourth thoracic vertebra. Many of the adjacent vertebrae are pneumatically connected with these two sacs, which also are connected with each other.

The anterior thoracic sac, as its name indicates, is in the anterior part of the thoracic cavity. It is related to the structures there and communicates with pneumatic cavities in the bones of the shoulder girdle, the humerus, the sternal ribs, and sternum. This sac has a right and left axillary diverticulum which projects into intermuscular spaces of the shoulder.

The posterior thoracic air sacs are paired. These are bounded by the pulmonary and thoraco-abdominal diaphragms and do not have connections with air cavities of bones (Bradley, 1938).

The lesser abdominal air sacs are paired. These laterally compressed spheroids are located on the anterior part of the abdominal wall at about its middle third. This pair also, according to Bradley (1938), does not make connections with cavities in bones.

The left is quite a bit the larger of the paired greater abdominal air sacs. These sacs are co-extensive with the abdominal cavity on each side of the

medially located viscera. There is division of opinion as to the pneumatization of the femur. Bradley (1938) states that the sacrum, hip bone, and femur all three have air connections with these sacs. In McLeod and Wagers' paper (1939) they stated they were not able to find connections of the greater abdominal air sacs with the femur, but in the discussion of the paper others had found the femur pneumatized. The writer was not able to find any evidence of pneumatic cavities in the femurs of a large number of birds which he studied.

### THE URINARY ORGANS

The kidneys present usually three lobes of unequal size. They are limited anteriorly by the lungs and extend posteriorly through the length of the pelvic cavity filling in the fossae formed by the pelvic bones and the vertebrae. The ureters may easily be traced from lobe to lobe and to the terminations in the dorsal wall of the urodaeum of the cloaca. Chauveau and Arloing (1891) state that the ostrich is the only bird possessing a urinary bladder. The finer structure of the kidney of the fowl resembles very closely that of the same mammalian organ.

### THE REPRODUCTIVE ORGANS

#### THE MALE GENITALIA

The testes are intra-abdominal organs in fowls. Each testis is ellipsoidal and is suspended by the mesorchium ventral to the anterior lobe of the corresponding kidney. From a diminutive epididymus, each vas deferens pursues a flexuous course posteriorly to enter the urodaeum through a papilla slightly lateral to the entrance of the ureter. In the chicken no penis is present. However, in the male goose, duck, and ostrich, a penis type of erectile organ may be found. The erectile tissue is composed of lymph spaces (Schauder, 1923). Accessory male genital organs are absent. The microscopic structure of the avian testis and vas deferens follows closely that for the same mammalian organs.

#### THE FEMALE GENITALIA

Only the ovary and the oviduct on the left side persist in most birds. The ovarian mass of loosely connected yolk follicles lies immediately posterior to the left lung. It is supported dorsally by a fold of peritoneum. Medially it is bounded by the intestine and its mesentery, dorsally by the body wall, and laterally by the left abdominal air sacs. Vermeulen (1929) says that in some birds of prey the right ovary persists but not the right oviduct. The ova from such a functioning ovary would have to enter the left oviduct. This is facilitated, he states, by the location of the right ovary in a position posterior to the left.

There is a limited amount of stroma in the ovary. In the stroma is considerable smooth muscle. The follicles are composed of a Theca folliculi in which may be seen an external portion (Theca externa) of fibers, and an inner portion (Theca interna), a more cellular layer. Deeply to this is the granulosa which has cuboidal to columnar cells. The vitelline membrane surrounding the yolk lies next to this layer of cells. The yolk contains the true ovum which becomes the blastoderm.

The newly laid egg is composed of various layers. These will be described going from the outside to the inside. The calcareous shell may be considered to have three strata: an outer homogenous, a middle fibrous, and a deep particulate. An outer thicker part and an inner thinner portion, both composed of interlacing organic strands, compose the shell membrane. At the larger end of the egg the two portions of the shell membrane are separated by an air space. The white of the egg, or albumen, which composes the greater part of the egg may be seen to have an outer, more fluid albumen, an inner denser albumen forming the greater part, and the chalazae which are coils of denser albumen extending from the yolk to the shell membranes at the poles of the egg. A structureless vitelline membrane surrounds the yolk. In the yolk may be seen alternating narrow light and wider dark bands of white and yellow yolk, respectively. These layers are somewhat more evident in the boiled egg. In one portion of the yolk a mass of white yolk extends from near the vitelline membrane to the center of the yolk. Its central portion is expanded. This mass of white yolk is called the latebra. At the point of its contact with the vitelline membrane there is a pale disc of cells called the blastoderm. The cells of the blastoderm give rise to the embryo. The yolk is made up of microscopic granules or spheres with but little interstitial fluid. The specific gravity of the parts of the yolk are such that shortly after turning, the blastodermal portion assumes a position on the upper side.

The oviduct of the fowl is a tortuous tube extending on the left side of the abdominal cavity from the ovary to the urodaeum into the cavity of which it empties. It is suspended by a dorsal peritoneal ligament which is attached to the abdominal roof, while the ventral border of the ventral ligament is free. Bradley (1938) says this ventral border contains muscle. The oviduct in the laying hen may be as much as 30 inches in length, according to Bradley (1938).

Five portions with fairly distinctive structure and function may be described. The serosal tunic is continuous with the dorsal and ventral ligaments. The muscular tunic is typically composed of an inner circular layer and an outer longitudinal. It is thin anteriorly but increases in thickness posteriorly. The submucosa and the propria of the mucosa are continuous. The mucosa is much folded throughout. Its epithelial layer is composed of columnar cells both of the ciliated and nonciliated types.

The infundibulum, which is the first part, is characterized by a very thin wall and many folds in the mucosa. It receives the ovum. The second portion, called the Pars albuminifera, or albumen-secreting region, is characterized by the density of its glandular region in the propria. The third portion, the isthmus, is similar in structure to the preceding region. Most of the albumen is laid down in the second and third segments and the shell membrane in the third portion. In the uterus (the fourth segment) which, too, has a glandular propria, the shell is formed. The final or fifth portion is termed the vagina. The mucosa of the vaginal portion presents many longitudinal folds, and the propria soon becomes free of glands. This portion of the oviduct apparently contributes little, if any, to the structure of the egg, serving simply as a passageway (Pearl and Curtis, 1912; Surface, 1912; Giersberg, 1923; Bradley, 1927, 1938; van den Broek, 1933; Asmundson and Burmester, 1936).

### THE ORGANS OF THE BLOOD AND LYMPH VASCULAR SYSTEM

The heart of birds is similar to that of mammals. According to Vermeulen (1929) it may equal as much as 25 per cent of the body weight in small rapid-flying birds, while in the chicken it equals 4 to 8 per cent, as compared to 1.5 to 1.7 per cent for man and large domestic animals.

The origin of the arteries carrying blood anteriorly is from the aorta, which first gives off a left brachio-cephalic and then a right brachio-cephalic artery. These give rise to their respective common carotids and subclavians. The latter give off large pectoral arteries and are continued by the brachial arteries to the wings. The common carotids in the neck run close together ventral to the longus colli muscle for most of the distance. The thoracic portion of the aorta beyond the origin of the right brachio-cephalic artery and the abdominal aorta gives off the ordinary segmental paired and the unpaired arteries. The external iliac arteries (s. femoral) are small. The ischiadic arteries are large arteries each passing through the sciatic foramen paralleling the sciatic nerve to supply pelvic and posterior-limb muscles. The internal iliac arteries (s. hypogastric) are small and supply the pelvic wall. The medial sacral artery is the direct continuation of the aorta.

The larger veins differ somewhat from those of mammals. The jugular veins are not satellites of the common carotids which are more medially located (see above). These veins parallel the trachea one on each lateral side. They always anastomose at the base of the cranium, according to Chauveau and Arloing (1891). The right jugular may be of a larger caliber than the left.

The jugulars unite with veins which are satellites of the branches of the brachio-cephalic arteries to form two similar trunks, called anterior venae cavae. These two, with the posterior vena cava, return the blood of the systemic circulation to the right atrium. The posterior vena cava receives hepatic
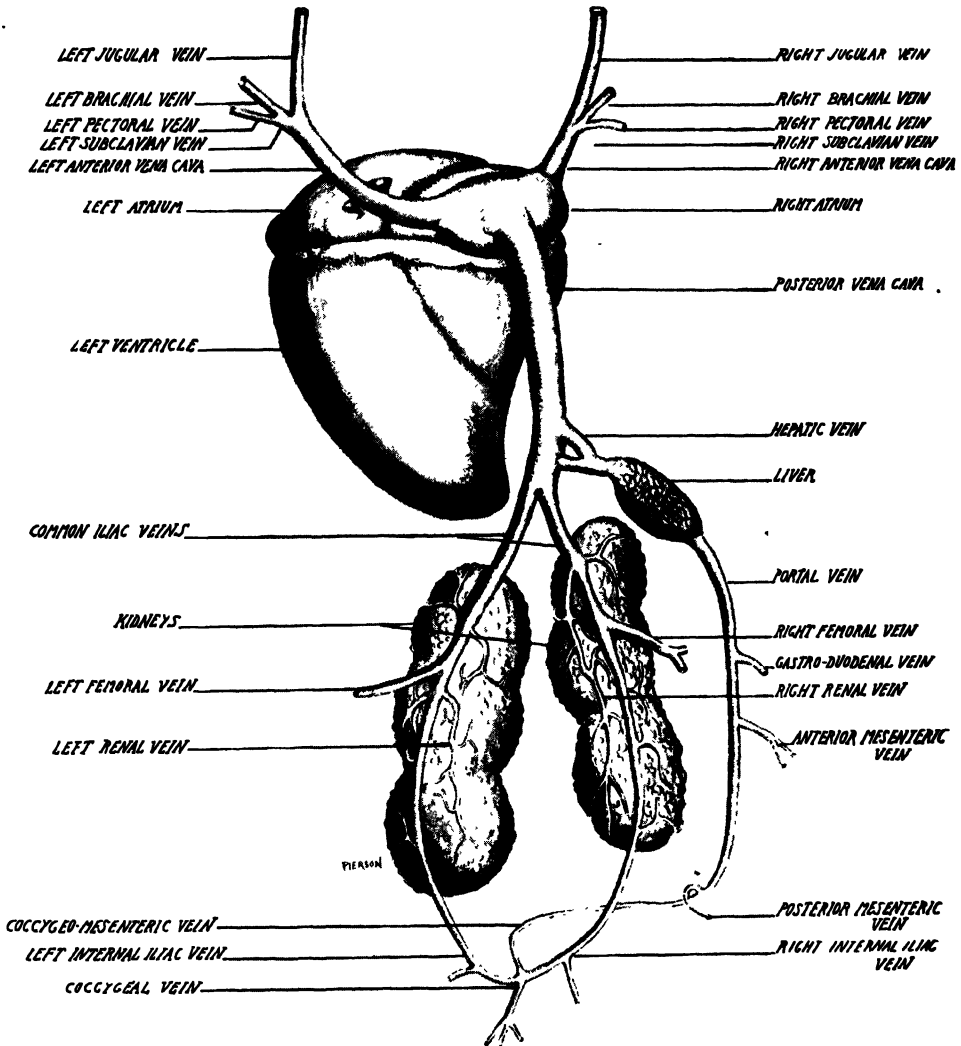
FIG. 1.6. Semischematic drawing of heart and veins of the common fowl. Pulmonary veins are partly covered by left anterior vena cava.

veins from the liver and two common iliac veins. The formation (Fig. 1.6) of these will now be discussed.

Coming from the tail the coccygeal vein sends an anastomosing branch to both internal iliac veins, and through the coccygeo-mesenteric vein joins the posterior mesenteric vein. The posterior mesenteric, the anterior mesenteric,

and the gastro-duodenal veins form the portal vein which is continuous through the hepatic vessels with the hepatic veins.

Each internal iliac vein drains the pelvic wall and, after receiving the aforementioned anastomosing branch from the coccygeal vein, passes anteriorly through the substance of the kidney, receiving the femoral vein at the junction of the anterior and middle lobes, and later the renal vein, to form a common iliac vein. The two common iliac veins unite to form the short posterior vena cava just anterior to the kidneys.

The pulmonary veins, one from each lung, enter the left atrium through a common opening.

### THE LYMPHATIC SYSTEM

Typical lymphatic nodules with germinal centers have been found by Jordan (1936, 1937), but in referring to nodules in the bone marrow of turkeys he wrote, "In agreement with conditions in chickens and pigeons only the marrow of young individuals contains nodules" (Jordan, 1937). Disselhorst, however, has been reported by Jacobshagen (1937) to have found no mitotic figures in germinal centers in lympho-reticular tissue in the wall of the Cloacae Fabricii. According to Fürther (1913), ducks and geese have cervico-thoracic and lumbar lymph nodes in which lymphoblastic follicles and germinal centers appear in the first month after hatching. At the end of the second month he found the lymph nodes fully developed and observed that a hilus was not present in the avian lymph node.

Lymph nodules were said by Bradley (1938) to be present throughout the entire intestine, often replacing parts of the mucous membrane.

Baum (1930) described two thoracic ducts, a right and a left, which he divided into thoracic and lumbar portions. Each of these ducts emptied into the Vena cava cranialis corresponding to its side. He also stated that the larger lymph vessels of the hen either drained into the thoracic ducts or directly into the venous system, as lymph nodes are not present in the chicken. Fürther (1913) described similar bilateral thoracic ducts in ducks and geese.

Dransfield (1945), in his studies, found that "Lymphatic glands are completely absent in the domestic fowl, and the author agrees with other authors that plexuses of the lymphatics appear to take the place of glands." Plexuses such as these were found especially on the course of the lymphatics of the abdominal viscera.

Valves, although few, are present in lymph vessels of birds (Drinker and Yoffey, 1941; Grau, in Ellenberger and Baum, 1943).

According to Drinker and Yoffey (1941), lymph hearts are no longer present in post-embryonic birds.

The sinuses in the avian lymph nodes are devoid of a reticulum but are

lined by endothelial cells (Baum and Trautmann, 1933; Drinker and Yoffey, 1941; Grau, in Ellenberger and Baum, 1943).

Cortical and medullary regions of nodules (Fig. 1.5) were observed by Calhoun (1933) in the wall of the bursa of Fabricius, although in many other regions of the digestive tube she found diffuse lymphoid tissue.

The spleen is small, brownish red, and roundish, oval or somewhat disc-shaped. It lies to the right of the glandular stomach.

### THE NERVOUS SYSTEM

The parts of the brain and spinal cord are quite similar to those of mammals. Of the fissures of the cerebrum, the lateral fissure of the cerebrum is the only one developed. The cerebellum is considerably reduced. The pons is absent. There are twelve cranial nerves as in mammals.

Further and detailed knowledge of the anatomy of the avian central nervous system may be found in the works of Stieda, 1869; Bumm, 1883; Turner, 1891· Boyce and Warrington, 1898, 1899; Münzer and Wiener, 1898; Edinger and Wallenberg, 1899; Edinger, 1903; Wallenberg, 1904; Kalischer, 1905; Lapicque and Girard, 1906; Schroeder, 1912; Dennler, 1922; Groebbels, 1924; Huber and Crosby, 1929; Papez, 1929; Sanders, 1929; Craigie, 1932; Kappers, Huber, and Crosby, 1936.

The spinal cord has the same two enlargements as in mammals. There is one at the cervico-thoracic junction and one in the lumbar region. Spinal nerves are given off intervertebrally as in mammals. Brachial and lumbo-sacral plexuses too, are similar to those of mammals. Of especial interest, may be mentioned a cutaneous branch of the femoral and one of the sciatic nerve. The femoral nerve gives off a large cutaneous branch which emerges about the middle of the posterior border of the sartorius muscle. Likewise, the sciatic nerve gives off a cutaneous branch which emerges at the proximal part of the middle third of the anterior border of the semitendinosus muscle. These two nerves innervate the skin in areas adjacent to their points of emergence.

The sympathetic trunks are similarly disposed as in mammals. Especially may be mentioned the location of the splanchnics. From the thoracic portion of the sympathetic trunk, there are given off at the third, fourth, and fifth intercostal spaces, branches which join at the celiac artery to form the greater splanchnic nerve. The lesser splanchnic is given off at the level of the adrenals and gonads.

### ORGANS OF SPECIAL SENSE
#### THE EYE

The lower eyelid is somewhat larger than the upper. Eyelashes are re-placed by fine feathers. A very extensive nictitating membrane is present.

Its base is medial. It is capable of covering the entire palpebral surface of the eyeball. Bradley (1938) states that a gland is present in the third eyelid. This glandular structure is commonly called the gland of Harder.

The structure of the wall of the eyeball differs somewhat from that in mammals. The sclera has an inner layer of hyaline cartilage, and a ring of bony plates is found at the corneo-scleral junction.

Projecting into the vitreous from an otherwise avascular retina is the pecten. It is a vascular and pigmented folded structure with its base on the optic disc. Duke-Elder (1934) gives a discussion of this structure with considerable review of literature.

Vermeulen (1929) states that the yellow color of the iris is due to the presence of fat.

### THE EAR

The auricula is absent. The external acoustic meatus is guarded by some small feathers in most birds. But one ossicle, termed the columella, spans the space between the tympanic membrane and the fenestra ovalis. The middle ear extends into air spaces in the basi-occipital and basi-sphenoid bones (Vermeulen, 1929). A slightly bent tube forms the cochlea.

### THE OLFACTORY ORGAN

The distribution of the olfactory nerves is, in general, similar to that for mammals.

### THE ORGAN OF TASTE

Owing to the horny nature of the dorsum of the tongue, taste papillae are absent. No mention, in the literature reviewed, is made of a chorda-tympanic branch of the facial nerve, and since there are said to be taste cells about the oral glands, base of tongue, and in the posterior portion of the palate (Vermeulen, 1929), these must belong to the glosso-pharyngeal nerve.

### THE ORGAN OF TOUCH AND STRUCTURE OF THE SKIN AND ITS APPENDAGES

Touch corpuscles are found in large numbers in various places in the skin. In the beak, in head and neck appendages, and in the wall of the cloaca they are especially plentiful (Vermeulen, 1929).

The structure of the skin is in general similar to that of mammals. Sudoriparous glands are lacking. Sebaceous glands, too, are wanting. There is, however, a large uropygial gland (oil gland) located dorsal to the last sacral vertebra. This is a bilobed gland of the sebaceous type. From a sinus in the center of each lobe the secretion passes by way of a tube to the outside through a single papilla common to the two lobes. It is most highly developed in waterfowl.

The skin of the metatarsal and of the digital regions of the foot presents

horny scales. Vermeulen (1929) says these scales, especially on the dorsal surfaces of the appendages, become harder and coarser with age. Many fowl have a spur on the medial side of the metatarsal region. Vermeulen (1929) states that it is a highly developed scale. It has a bony process on the metatarsal bone for a core. Its horny covering appears to grow from base to apex. The size of the claws varies from rudimentary in waterfowl to well-developed structures in birds of prey. Pads form cushions on the plantar sides of the digits for the absorption of shock. Webs between the digits vary in extent in different waterfowl.

In the deeper layers of the skin, there is considerable muscle which is attached to the follicles of the feathers.

The comb of fowls has a somewhat lanceolate core of adipose tissue which is narrowed posteriorly. Thin, dense connective tissue laminae separate this core from a layer of muco-elastic tissue. This latter layer is composed of large anastomosing cells with a fine reticulum in their cytoplasm. In the meshes there is a clear mucus slightly stainable by ordinary methods but specifically stained by mucicarmine and mucihematin. There are many elastic fibers which with collagenous fibers extend from the laminae to the subepithelial zone of connective tissue. Large blood vessels of the core are connected by capillaries with a great number of sinusoidal vessels in the subepidermal connective tissue.

There is a well-developed nerve supply with many pacinian corpuscles. The epidermis is a stratified epithelium. The points of the comb have a structure like that of the comb proper.

The cone-shaped comb of the turkey has large fasciculi of smooth muscle around a dense connective tissue core. This core contains erectile tissue. Its arteries are medium-sized with heavy muscular walls. The subepithelial sinusoids are sparse while the subepithelial arteries are tortuous. The muco-elastic layer may be absent.

The helmet of the guinea fowl has a core of spongy bone with areolae into which extend diverticula from air sacs.

Wattles have a structure somewhat similar to that of the comb. The fatty core may be reduced or interrupted, and the muco-elastic layer may be more hyalin in character.

The muco-elastic layer of the comb of the cock disappears after complete castration. The description of the comb and wattles follows that of Champy and Kritch (1926).

Feathers are epidermal structures similar in that respect to hair, and also similar in that the proximal end is in a follicle.

A typical feather (Fig. 1.7) has a tubular shaft or axis. The proximal portion of the shaft is called the quill or calamus. It is a cornified, somewhat translucent tubule, the lumen of which contains cellular debris and air

Area magnified in (B)

Vane or vexillum

Groove

Rachis

Hyporachis

Distal umbilicus

Quill or calamus

Proximal umbilicus

A.

Barb

Curved end of
proximal barbule

Proximal barbule

Distal barbule

Hooklet

Rachis

Barb

B.
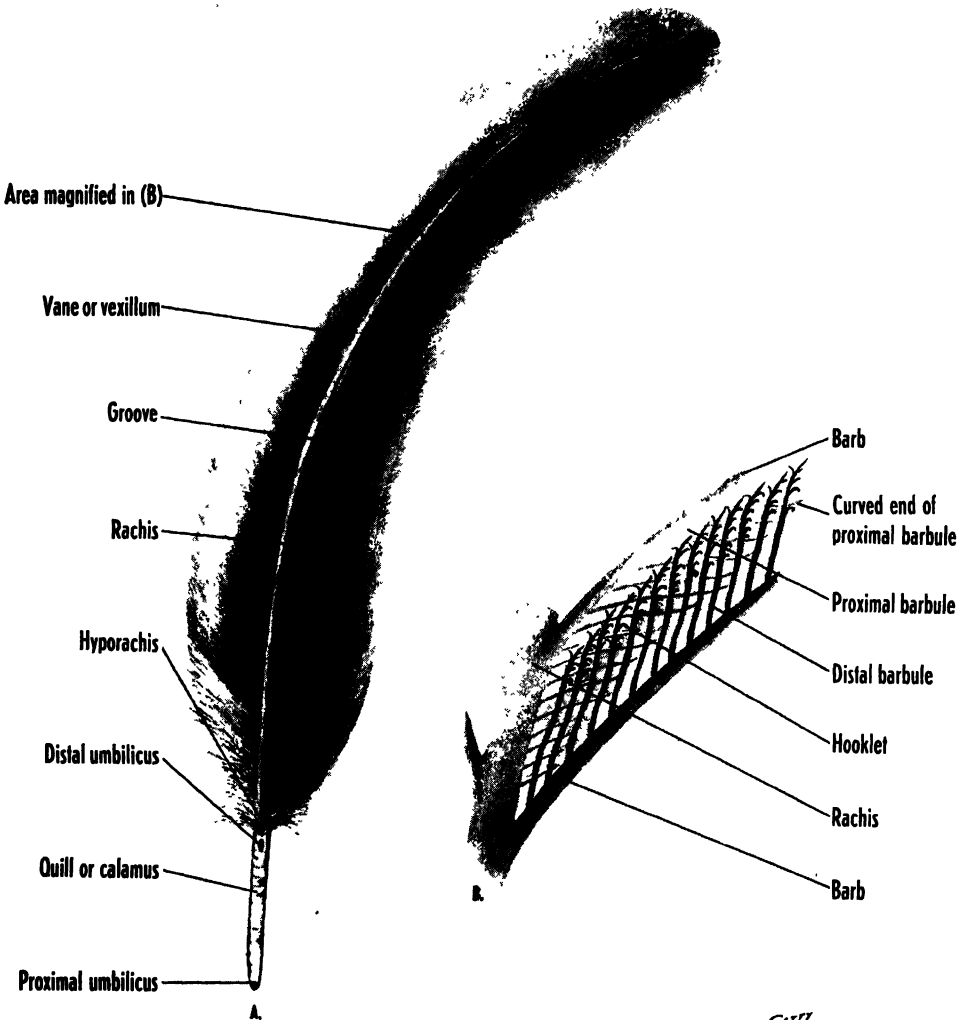
FIG. 1.7. A—Deep (concave) surface of typical flight feather. B—Section of feather "A"
viewed from superficial (convex) surface.

spaces. The follicular end presents an opening, the proximal umbilicus, into
which a dermal papilla projects. At the distal part of the quill, there is a
foramen called the distal umbilicus. Closely associated with it and projecting
from the shaft frequently is a small feather termed the hyporachis.

Distal to the quill or calamus is the part of the shaft called the rachis. It
is groved on the side facing the body. This portion with its two rows of
barbs forms the vane or vexillum. The barbs are lamellae which arise from

the rachis and are directed obliquely distally. In many feathers each barb has a distal row of barbules which have several hooklets near their terminal ends. The distal row of barbules overlaps a row of proximal barbules located on the succeeding barb. The barbules of this proximal row are curved. The concavity of the curve lies immediately deeply to the ends of the distal row of barbules (Fig. 1.7B). This interlocking device makes of the vane an elastic membrane providing resistance to air during flying. In downy types of feathers barbules may be partly or wholly absent. The shaft, too, may be considerably reduced. Molting is accompanied by the formation of a new feather which arises in the follicle already present and pushes out the old feather.

As is the case of the distribution of hair in mammals, feathers are arranged in areas and lines which are termed pterylae. The term, apteria, is applied to the intervening bare areas.

## THE ENDOCRINE ORGANS

*Pancreas.* Reference was made to the islets of Langerhans in the description of the pancreas in the section devoted to the digestive system.

The *thyroid glands* of birds consist of two elliptical lobules. These lie intrathoracic on the large trunks of the vessels arising directly from the heart. The structure of the avian thyroid is very similar to that found in mammals.

A *parathyroid* lobule is found at the posterior pole of each thyroid gland. Immediately posterior to this is a smaller second parathyroid lobe. The structure of the parathyroid of fowl is very similar to that of mammals.

The *thymus* is composed of many lobules which may extend throughout the cervical region. Its structure and involution resemble the same for mammals.

The *hypophysis cerebri* varies somewhat in structure from that found in mammals. A Pars intermedia cannot be differentiated. The Pars anterior, cytologically, has two distinct regions which Rahn and Painter (1941) term "cephalic" and "caudal" lobes. These lobes are divided by a line which begins dorsally at the base of the Pars tuberalis which is located on "the posterior portion of the cephalic lobe." It extends ventrally to "remnants, if any, of the hypophyseal stalk." Embryological verification was made of the position of the two portions. They (Rahn and Painter, 1941) found the cephalic lobe to contain chromophobes, light staining acidophils, and basophils; and in the caudal lobe, which lies "nearest to the infundibular process, chromophobes, basophils, and deep staining, coarsely granular acidophils." These acidophils with large granules were considered to be distinct from the acidophils of the cephalic lobe. This caudal lobe was said to resemble the Pars anterior of mammals.

The Pars tuberalis extends from the optic chiasma caudally and surrounds the infundibular stalk.

The cavity of the infundibulum is present in the adult. Rahn and Painter (1941) based their findings upon studies of the pituitary of the chicken and duck.

The Pars nervosa is separated in the mature fowl from the Pars anterior by a sheet of connective tissue (Atwell, 1939; Economo, 1899).

## REFERENCES

Arey, L. B.: 1946. Developmental Anatomy. Part III. A laboratory manual of embryology. Chap. XXII. The study of chick embryos. W. B. Saunders, Philadelphia. P. 508.

Asmundson, V. S., and Burmester, B. R.: 1936. The secretory activity of the parts of the hen's oviduct. Jour. Exper. Zool. 72:225.

Atwell, W. J.: 1939. The morphogenesis of the hypophysis cerebri of the domestic fowl during the second and third weeks of incubation. Anat. Record 73:57.

Bailey, F. R., and Miller, A. M.: 1929. Textbook of Embryology. Wm. Wood & Co., New York. Chapter VI, p. 66.

Baum, H.: 1930. Das Lymphgefässsystem des Huhnes. Julius Springer, Berlin.

———— and Trautmann, A.: 1933. (g) Vögel. VII. Lymphgefässsystem. Gefässsystem. Bolk, L., et al. Handbuch d. vergl. Anat. d. Wirbeltiere. Urban & Schwarzenberg, Berlin. 6:843.

Beams, H. W., and Meyer, R. K.: 1931. The formation of pigeon "milk." Physiol. Zool. 4:486.

Boyce, R., and Warrington, W. B.: 1898, 1899. Observations on the anatomy, physiology, and degenerations of the nervous system of the bird. Proc. Roy. Soc., London 64:176. See also Phil. Tr. of the Roy. Soc., London, Ser. B 191:293.

Bradley, O. C.: 1927. Notes on the histology of the oviduct of the domestic hen. Jour. Anat. 62:339.

————: 1938. The Structure of the Fowl. Oliver & Boyd, Edinburgh.

Bumm, A.: 1883. Das Grosshirn der Vögel. Zeitschr. f. wiss. Zool., 38:430.

Calhoun, M. L.: 1933. The microscopic anatomy of the digestive tract of Gallus domesticus. Ia. St. Coll. Jour. Sci. 7:261.

Chamberlain, F. W.: 1943. Atlas of avian anatomy, osteology, arthrology, myology. Mich. St. Coll. Agr. Exper. Sta.

Champy, C.: 1934. Manuel d'embryologie. Masson & Co. Paris.

———— and Kritch, N.: 1926. Étude histologique de la crête des gallinacés et de ses variations sous l'influence des facteurs sexuels. Arch. de Morph. Gen. et Exper. No. 25:1.

Chauveau, A., and Arloing, S.: 1891. The Comparative Anatomy of the Domesticated Animals. (Second English ed., Trans. by Geo. Fleming). D. Appleton & Co., New York.

Cholodkowsky, N.: 1892. Zur Kenntnis der Speicheldrüsen der Vögel. Zool. Anz. 15:250.

Craigie, E. H.: 1932. The cell structure of the cerebral hemisphere of the humming bird. Jour. Comp. Neurol. 56:135.

Cuvier, G. L. C. F. D.: 1832. Extrait d'un mémoire sur les progrès de l'ossification dans le sternum des oiseaux. Ann. des Sci. Nat. 25:260.

Dennler, G.: 1922. Zur Morphologie des Vorderhirns der Vögel. Der Sagittalwulst. Folia Neurobiol. 12:343.

Dransfield, J. W.: 1945. The lymphatic system of the domestic fowl. Vet. Jour. 101:171.

Drinker, C. K., and Yoffey, J. M.: 1941. Lymphatics, Lymph, and Lymphoid Tissue. Harvard Univ. Press.

Duke-Elder, W. W.: 1934. Textbook of Ophthalmology. Vol. I. C. V. Mosby Co., St. Louis.

Duval, M. M.: 1889. Atlas d'embryologie. Masson, Paris.

Economo, C. J.: 1899. Zur Entwicklung der Vogelhypophyse. Sitzungsberichte d. Akad. d. Wiss. 108. Abt. III:281.

Edinger, L.: 1903. Untersuchungen über die vergleichenden Anatomie des Gehirns. 5. Das Vorderhirn der Vögel. Abhandl. d. Senckenb. nat. Gesellsch., Frankfurt am Main, 20, Heft. 4:343.

———— and Wallenberg, A.: 1899. Untersuchungen über das Gehirn der Tauben. Anat. Anz. 15:245.

(Ellenberger-Baum), Zietzschmann, O., Ackerknecht, E., Grau, H.: 1943. Handbuch vergl. Anat. Haustiere. Springer, Berlin.

Fischer, G.: 1905. Vergleichend-anatomische Untersuchungen über den Bronchialbaum der Vögel. Zoologica. 19, Heft 45:1.

Foster, M., Balfour, F. M., Sedgwick, A., and Heape, W.: 1902. The Elements of Embryology. Part I. The history of the chick. Macmillan & Co., Ltd., London. Pp. 1–303.

Fürther, H.: 1913. Beiträge zur kenntnis der Vogellymphknoten. Jenaische Zeitschr. Natur-wissensch. 50:359.

Giersberg, H.: 1923. Untersuchungen über Physiologie und Histologie des Eileiters der Reptilien und Vögel; (nebst einen Beitrag zur Fraser-genese). Zeitschr. wiss. Zool. 120:1.

Groebbels, F.: 1924. Untersuchungen über den Thalamus und das Mittelhirn der Vögel. Anat. Anz. 57:385.

Heidrich, K.: 1905. Mundhöhlenschleimhaut und ihre Drüsen bei *Gallus domesticus*. Diss. Giessen. Published in 1908 as Die Mund-Schlundkopfhöhle der Vögel und ihre Drüsen in Gegenbaur Morph. Jahrb. 37:10.

His, W.: 1868. Untersuchungen über die erste Anlage des Wirbelthierleibes. Die erste Entwickel-ung des Hühnchens im Ei. F. C. W. Vogel. Leipzig.

Huber, G. C., and Crosby, E. C.: 1929. The nuclei and fiber paths of the avian diencephalon, with consideration of telencephalic and certain mesencephalic centers and connections. Jour. Comp. Neurol. 48:1.

Huetner, A. F.: 1941. Fundamentals of Comparative Embryology of the Vertebrates. Chapters IX–XV. The embryology of the chick. Macmillan Co., New York. P. 192.

Jacobshagen, E.: 1937. (c) Vögel, IV. Mittel-und Enddarm, (Rumpfsdarm), Darmsystem und Atmungssystem der Kranioten. Bolk, L., *et al.* Handbuch d. vergl. Anat. d. Wirbeltiere. Urban & Schwarzenberg, Berlin, 3:654–71.

Jordan, H. E.: 1936. The relation of lymphoid tissues to the process of blood production in avian bone marrow. Am. Jour. Anat. 59:249.

———: 1937. The relation of lymphoid nodules to blood production in the bone marrow of the turkey. Anat. Record 68:253.

Kalischer, O.: 1905. Das Grosshirn der Papageien in anatomischer und physiologischer Beziehung. Abhandl. d. kön. preuss. Akad. d. Wissensch., Berlin. 4:1.

Kappers, C. U. A., Huber, G. C., and Crosby, E. C.: 1936. The Comparative Anatomy of the Nervous System of Vertebrates Including Man. Macmillan Co., New York.

Keibel, F.: 1900. Normentafeln zur Entwicklungsgeschichte der Wirbelthiere. II. Keibel, F., und Abraham, K. 1900. Normentafel zur Entwicklungsgeschichte des Huhnes *(Gallus domesticus)*. Gustav Fischer, Jena. Pp. 1–132.

Kerr, J. G.: 1919. Textbook of Embryology. Vol. II. Vertebrata (except mammalia). Chap. X. The practical study of the embryology of the common fowl. Macmillan & Co., Ltd., London. Pp. 509–57.

Lapicque, L., and Girard, P.: 1906. Poids des diverses parties de l'encéphale chez les oiseaux. Compt. rend. Soc. de biol. 61:30.

Leasure, E. E., and Link, R. P.: 1940. Studies on the saliva of the hen. Poultry Sci. 19:131.

Lillie, F. R.: 1918. The Development of the Chick. Henry Holt & Co., New York.

McEwen, R. S.: 1931. Vertebrate Embryology. Part IV. The development of the chick. Henry Holt & Co., New York. Pp. 291–472.

McLeod, W. M., and Wagers, R. P.: 1939. The respiratory system of the chicken. Jour. Am. Vet. Med. Assn. 95:59.

Minot, C. S.: 1903. A Laboratory Textbook of Embryology. Chap. V. Study of young chick embryos. P. Blakiston's Son & Co., Philadelphia. Pp. 269–305.

Münzer, E., and Wiener, H.: 1898. Beiträge zur Anatomie und Physiologie des Centralnerven-systems der Taube. Monatschr. f. Psychiat. u. Neurol. 3:379.

Needham, J.: 1931. Chemical Embryology. Macmillan Co., New York.

Papez, J. W.: 1929. Comparative Neurology. T. Y. Crowell Co., New York.

Patten, B. M.: 1946. The embryology of the chick. P. Blakiston's Son & Co., Inc., Philadelphia.

Pearl, R., and Curtis, M. R.: 1912. Studies on the physiology of reproduction in the domestic fowl. V. Data regarding the physiology of the oviduct. Jour. Exper. Zool. 12:99.

Rahn, H., and Painter, B. T.: 1941. A comparative histology of the bird pituitary. Anat. Record 79:297.

Sanders, E. B.: 1929. A consideration of certain bulbar, midbrain, and cerebellar centers and fiber tracts in birds. Jour. Comp. Neurol. 49:155.

Schauder, W.: 1923. Anatomie der Hausvögel. Martin, P. Lehrbuch der Anatomie der Haustiere. Schickhardt & Ebner, Stuttgart. 4:339–98.

Schroeder, K.: 1912. Der Faserverlauf im Vorderhirn des Huhnes, dargestellt auf Grund von entwicklungsgeschichtlichen (myelogenetischen) Untersuchungen, nebst Beobachtungen über die Bildungsweise und Entwicklungsrichtung der Markscheiden. J. f. Psychol. u. Neurol. 18:115.

Stieda, L.: 1869. Studien über das zentrale Nervensystem der Vögel und Säugetiere. Zeitschr. f. wissensch. Zool. 19:1.

Surface, F. M.: 1912. Histology of the oviduct of the domestic hen. Maine Agr. Exper. Sta. Bul. 206:395.

Turner, C. H.: 1891. Morphology of the avian brain. I. Taxonomic value of the avian brain and the histology of the cerebrum. Jour. Comp. Neurol. 1:39.

van den Broek, A. J. P.: 1933. (f) Vögel. Gonaden und Ausführungsgänge. II. Geschlechtorgane. Urogenitalsystem. Bolk, L., et al. Handbuch d. vergl. Anat. d. Wirbeltiere. Urban & Schwarzenberg, Berlin. 6:94.

Venzke, W. G.: 1942. The embryological development and physiology of the endocrine organs of the common fowl (Gallus domesticus). Thesis. Ia. St. Coll.

Wallenberg, A.: 1904. Neue Untersuchungen über den Hirnstamm der Taube. Anat. Anz. 24:142–55, 357–69.

Vermeulen, H. A.: 1929. Anatomie und Physiologie des Geflügels. Handbuch der Geflügelkrankheiten und der Geflügelzucht. T. van Heelsbergen. Ferdinand Enke, Stuttgart.

# CHAPTER TWO

## DIGESTION

*By* E. A. Hewiti, *Department of Veterinary Physiology and Pharmacology,*
*Iowa State College, Ames, Iowa*

☆　　☆　　☆

The alimentary tract of birds differs to a considerable degree from that of other animals. There is no provision for mastication in the mouth because of the absence of teeth. The "egg tooth" of young birds is a small horny point on the upper beak which soon disappears. The edges of the beak are smooth in birds that simply pick up their food; however, they are ribbed and serrated in those birds that break up their food before swallowing. The hard palate contains tactile corpuscles. The mucous membrane contains no glands, but there are numerous glands in the submucosa especially on the roof of the pharynx. The secretion into the pharynx is fairly profuse. The base of the tongue is supported by a small bone, the Os entoglossum. It is connected with the hyoid bone by a small joint. The structure of the hyoid is very complicated. By this structure quick movements of the tongue are made possible.

The food is swallowed almost immediately when it is taken into the mouth. The act of deglutition in birds differs in some respects from that of mammals. The lingual muscles are very poorly developed, except in the parrot. Owing to the rather rigid nature of the structures in the floor of the mouth and due to the absence of a mylohyoid muscle, it is impossible for swallowing to receive the muscular aid that it does in mammals. The bolus, therefore, instead of being pushed or shot into and through the pharynx by a contraction of the mylohyoid and other muscles, is propelled backward by a raising of the head and by giving it a quick backward thrust. It is of interest to note that when the mylohyoid muscle is paralyzed in mammals, these animals likewise raise the head in swallowing. Investigators have found that the food, regardless of its consistency, is carried down the esophagus by peristalsis at an average rate of 1.5 centimeters per second. There is no evidence of squirting or shooting of the food down the esophagus as occurs with liquid or semiliquid food in some mammals.

Although Calhoun (1933) found only mucous cells in these glands, Vermeulen (1929) reported that in some granivorous birds, serous cells

were present which produced a diastatic ferment. Leasure and Link (1940) found amylase in the salivary gland of the hen.

## CROP

The crop functions as a reservoir in which the food is stored and softened as long as the glandular stomach is in action. In the chicken the mucous membrane of the crop contains a few mucous glands near the esophageal inlet; the secretion from these is fairly profuse. Some investigators claim that no digestive enzymes are secreted by the crop while others claim that an amylase is produced by the gland cells which are confined to an area close to the junction with the esophagus. Formerly it was believed that gastric juice was regurgitated to the crop and that the food was partially digested there. Such a condition probably occurs only under pathological conditions. A few birds feed their young with the crop contents and can execute arbitrary vomiting movements. During this time of infant nursing, the mother pigeon secretes the so-called crop milk. This consists of fatty, degenerated, cast off exfoliated epithelial cells. The epithelial cells hypertrophy considerably for this purpose. The thickness of the epithelium of the crop in pigeons at ordinary times is about 0.16 mm., but at the time the eggs are hatched the cells may increase in thickness so that the epithelium may increase to a depth of 3 mm. in the mother pigeon. It is rather peculiar that male pigeons also may prepare an acid crop milk in which colostrum-like corpuscles may occur, which are not present in the product of the female pigeon.

The length of time that food remains in the crop varies mainly with the degree of filling of the organ. Browne (1922) found that if the different foods fed were of the same consistency, there was little or no mixing in the crop and that the foods left the crop in the order in which they entered. However, if foods having different consistencies were fed the softer food left the crop first. The greater the degree of filling, the slower on the average did the food pass on from the crop. Two main types of movements are characteristic of the crop; one type having to do with the forcing of ingesta into the proventriculus, and the other type associated with hunger. The movements concerned with forcing the food on are shallower and less energetic and also less rhythmic when the organ is full. In regard to the hunger contractions many observations indicate that these may be observed shortly after eating. These hunger contractions probably occur as soon as there is any accommodation for food in the crop. It is stated that these hunger contractions begin 30 to 45 minutes after eating. The contractions gradually increase in frequency and vigor until in 5 to 6 hours they occur in groups of six to twelve or more, separated by intervals of comparative rest. When the crop is well distended by food only occasional contractions can be detected by the balloon method. An hour or so after eating, periodic groups of hunger con-

tractions begin. These gradually become augmented, and the motility becomes marked several hours after eating. It appears, therefore, that hunger contractions in birds, occurring in the crop, are similar to those occurring in the stomach of mammals. The contractions begin, however, when there is relatively more food present in the crop of birds than in the stomach of mammals.

### PROVENTRICULUS

The proventriculus or glandular stomach is a small, fusiform organ situated between the two lobes of the liver anterior to the gizzard and at the level of the two anterior poles of the kidneys. In the wall of the proventriculus there occur two layers of glands, a superficial layer of simple tubular glands (mother cells) and a much heavier deep layer with compound tubular glands (covering cells). The excretory ducts of the deeper glands open on the surface between the superficial glands. Mucous cells are found among the epithelial cells of the superficial glands, and lymphoid tissue is located between the superficial tubules. This corresponds with the fundus region of the mammalian stomach. The glands of the proventriculus secrete gastric juice. The juice is similar in composition to the gastric juice of mammals; pepsin, rennin, and hydrochloric acid being present. The regulation of the secretion of gastric juice in birds is probably similar to that of mammals. Several investigators report that chemical stimuli also play a part in the regulation of secretion. The peptic ferment of the glandular stomach in many birds of prey can digest bones completely; other birds regurgitate such material. Many other indigestible materials such as cellulose, chitin, wool, and hair are frequently regurgitated by birds.

Owing to the small size of the cavity of the proventriculus the food does not remain in this organ long; therefore, the amount of gastric digestion taking place in the proventriculus must be inconsequential. It has been pointed out that the relatively dry nature of the gizzard contents (average moisture content 44.2 per cent) makes it improbable that any considerable amount of enzymatic action goes on in that organ. These facts lend support to the idea that the duodenum is the probable seat of the main action of gastric juice. The reaction of the duodenum is acid. The alkaline pancreatic juice and bile enter the duodenum near the posterior extremity of the duodenum. The point of entry of the bile and pancreatic juice is taken as the demarcation between the duodenum and ileum.

### GIZZARD

The gizzard or muscular stomach lies posterior to the liver. The segment of the alimentary tract between the proventriculus and gizzard varies in length in different species. In the chicken, it is longer than in the duck and pigeon. The gizzard is conspicuous by its biconvex shape and the bright

tendinous sheets with which both surfaces are covered. The deep glands of the proventriculus end abruptly and are followed by an increase in the length of the superficial tubular glands. These soon take on the characteristic aspect of the gizzard glands, and a keratinized layer formed from an exudate or secretion of the glands appears above them. This keratinized or horny layer forms the internal lining of the gizzard and is about three-fourths as thick as the glandular layer adjacent to it.

The mucosa is very rich in glands and corresponds with the pyloric part of the mammalian stomach. The gizzard is strongly developed especially in birds that eat grain. In chickens, turkeys, parrots, ostriches, and aquatic birds, it is considerably larger than the glandular stomach. In birds of prey and birds subsisting on fruit, it is less developed. That the development depends on the kind of feeding is evidenced from experimental results of feeding geese meat balls and flesh meal in which case the gizzard develops less in size than in geese fed on grain. The gizzard has a mechanical function, and the muscle walls in contracting are able to exert considerable force. Little stones are polished to remarkable smoothness. These stones facilitate the mechanical work and also exert a useful reflex stimulus. The stones may be discharged through the mouth as well as through the anus. Usually they are voided as very fine dust. Little if any digestion takes place in the gizzard, although a slight action might be exerted as the gastric juice secreted from the proventriculus is being mixed with the food.

### INTESTINAL TRACT

The intestinal tract is short in birds of prey. In chickens, pigeons, ducks, and geese it is four to six times the length of the body, and in the ostrich it is about eight times as long as the body. It is divided into four parts: the duodenum, ileum, ceca, and rectum. The wall of the entire length of the intestinal tract from the pylorus to the cloaca is marked by the presence of villi (Villi intestinales) and glands of Lieberkühn. It also contains much lymphoid tissue. Brunner's glands are absent. The duodenum forms a long loop which extends toward the pelvis. Between the sheets of mesentery connecting the two sides of the loop lies the pancreas. Posterior to the pylorus are located glands that resemble those found in the gizzard. Calhoun (1933) stated that this portion is similar to the mammalian duodenum. The secretion of these glands does not coagulate as does that of the gizzard; therefore, these glands may be considered as real pyloric glands.

The bursa of Fabricius opens into the dorsal cloacal wall. It is epithelial in origin, and in the young animal is composed of lymphoid tissue. It is composed of a cortical structure which consists of vascular connective tissue and lymphocytes and a medullary structure in which lymph-follicles are found. The bursa of Fabricius has been called a cloacal thymus. As soon as

puberty starts, this structure atrophies and then develops a thick fibrous wall. Its function is not understood.

## LIVER

The liver is the largest gland in the body and plays a very important part in general body nutrition. It may be classed as a peculiar form of a compound tubular gland whose cells resemble the serous secreting type. Roughly the liver contains about 25 per cent of the blood of the body. Its functions are various and depend upon the properties of the liver cell which constitutes the anatomical and physiological unit of the organ although the reticulo-endothelial cells of the liver capillaries apparently have a contributing part in certain functions, for example, in the destruction of red blood corpuscles which precede the formation of bile pigments. Such cells possess marked phagocytic properties.

It is stated that 30 per cent of the blood supply of the liver comes from the hepatic artery and 60 per cent from the portal vein. This portal blood is peculiar in that it has already passed through the capillaries of the digestive tract before entering the liver. The arterial supply is relatively meager and supplies the connective tissue framework intermingling with the portal blood at the periphery of the lobule. The source of the blood supply through the liver may be indicated by the following table:

| | |
|---|---|
| 1. Portal vein | 1. Hepatic artery |
| 2. Interlobular veins | 2. Interlobular arteries |
| 3. Branches to lobule | 3. Branches and capillaries in Glisson's capsule |

4. Intralobular capillaries
5. Central vein (intralobular)
6. Sublobular veins
7. Hepatic veins
8. Posterior vena cava

Each liver lobule also gives rise to bile capillaries which arise between the cells and carry off the bile produced by the liver cells. The bile capillaries occur as secretory canaliculi between the opposed surfaces of the hepatic cells. They are true secretory canaliculi by which the bile after secretion by the hepatic cells is collected and passed into a minute bile duct. The hepatic cells are pyramidal cells with the apex bordering the lumen of the tubule. A large spherical nucleus is located in the distal half of the cell. Coarse granules are found in the protoplasm which are undoubtedly glycogen granules as may be shown by color reaction. Fat globules occur in the hepatic cells and appear to be a normal constituent. They are small but increase in amount during fat digestion.

The hepatic cells also contain brown or yellowish-brown granules of

ferrugenous pigment usually more prone to occur in the cells in the interior of the lobule near the central vein.

The physiology of the liver naturally falls into two parts: one dealing with the composition, formation, and physiological significance of the bile; the other with the metabolic changes produced in the mixed blood of the portal vein and the hepatic artery passing through the lobules. In this latter division there is a decided difference in the function of the liver of birds and that of mammals. In mammals the formation of urea is very prominent, and uric acid is destroyed to a considerable degree, whereas in birds uric acid represents the chief end product of nitrogen metabolism. The glycogenic function is probably similar in the two kinds of animals. The liver has a number of important functions which may be summarized as follows:

1. The formation and secretion of bile

2. The formation and storage of glycogen and the regulation of the glucose content of the systemic circulation

3. The deamination of amino acids

4. The desaturation of fatty acids subsequent to their utilization by the tissues

5. The detoxication of poisonous substances brought to it by the blood

6. The aiding in the destruction of erythrocytes

The liver possesses other functions somewhat imperfectly understood at present. Some of these are, for example, the relation to the production of certain blood constituents, the formation of hemoglobin, and the detoxication of various organic and inorganic poisons.

Glycogen apparently constitutes a temporary reserve supply of carbohydrate material stored during digestion and utilized as a source of energy between digestion periods. As the blood of the portal circulation passes through the liver, the excess of sugar is abstracted by the liver and by a process of dehydration is converted to glycogen and retained temporarily.

The great importance of the formation of glycogen and the consequent conversion of the sugar supply of the tissues is evident when we consider the nutritive value of carbohydrate food. Carbohydrates form the bulk of the usual diet, and the proper regulation of the supply to the tissues is, therefore, of vital importance in the maintenance of a normal condition.

The blood sugar concentration varies in species as follows:

| | |
|---|---|
| Chicken | 0.139 |
| Human | 0.08–0.18 |
| Horse | 0.102 |
| Dog | 0.182 |
| Cow | 0.046 |
| Goat | 0.059 |
| Pig | 0.09 |

The stored glycogen is reconverted to dextrose by the liver enzyme, glyco-genase, and passed out to the blood as needed, the control of the output likely being of a hormone nature. The complete story is not known, but as part of it at least, attention has been called to the regulating action of three hormones, epinephrin, insulin, and the so-called diabetogenic hormone of the anterior lobe of the pituitary. Epinephrin in the blood stimulates glyco-genolysis, while insulin has the opposite effect. The part played by the anterior pituitary is a matter of speculation. According to one theory it is essential for the mobilizing action of epinephrin. It has been shown that under the influence of strong emotions, there is an increased secretion of epinephrin and a corresponding rise in the sugar of the blood. The value of this adaptation is supposed to lie in the fact that a larger supply of oxidizable material is thus placed at the disposal of the muscles at a time when there is need of greater muscular activity.

Some of the sugar of the blood formed from the glycogen, when an excess is eaten beyond the energy needs of the tissues, may be converted into fat and stored in the adipose tissues instead of being burned, and in this way it may be retained in the body as a reserve supply of food of a more stable character. The glycogen derived from protein foods, once it is formed in the liver has, of course, the same functions to fulfill.

Total removal of the liver is always followed by a decrease in blood sugar, and a definite and characteristic group of symptoms related to this decrease in the blood sugar develop. The intravenous administration of glucose after these symptoms have developed immediately alleviates the symptoms.

The terms glycogenesis, glycogenolysis, and glycolysis are used to desig-nate the formation of glycogen, its reconversion to dextrose, and ultimate utilization by the tissues, respectively. Glycogen is found in the body in tissues other than the liver. It is estimated to occur in resting muscle to the extent of 0.5 to 0.9 per cent, which would mean that muscle may store glyco-gen in an amount equal to the liver and further possesses a glycogenetic function. Muscle glycogen must be regarded as a temporary reserve supply of readily available material to meet the energy requirements in the very tissue that is the greatest user of energy. The rapidly growing tissues of the embryo contain a considerable amount of glycogen, and it would seem that the glycogenetic function may be exerted by quite a wide range of active tissues in the body. Glycogen reserves are quickly affected by conditions calling for increased metabolism in the body (muscular exercise, for example).

## PANCREAS

The pancreas is described, histologically, as a compound tubulo-alveolar gland. The cells lining the secreting portion of tubules belong to the serous

or albuminous type. The pancreatic secretion is a clear, alkaline liquid which coagulates on heating. The existence of secretory nerve fibers to the pancreas, the fibers of which run chiefly in the vagi and to a lesser extent in the splanchnics, was demonstrated by Pavlov and his co-workers. The secretion obtained by artificial stimulation of the nerves is characterized by a long latent period of some minutes. A chemical excitant (hormone) is also involved. Acids brought into contact with the mucous membrane of the duodenum set up a prompt secretion of pancreatic juice. This is explained on the basis that a special substance, "secretin," is formed by the action of the acid on prosecretin present in the mucous membrane of the small intestine; this is absorbed by the blood, carried to the pancreas, and stimulates the flow of pancreatic secretion. Secretin is not an enzyme since its activity is not destroyed by boiling. Bayliss and Starling (1902) discovered this substance and designated it as a hormone. This marks the discovery of such chemical substances which are now recognized as playing such vital parts in the regulation (humoral) of many vital functional activities. Secretin is also believed to stimulate the secretion of bile.

According to the evidence at present in our possession, we must believe that the pancreatic secretion consists of two parts:

1. A nervous secretion caused by secretory fibers in the vagi and splanchnics.

2. A chemical secretion due to the hormone, secretin.

These two secretions are different in character. The nervous secretion is thick, opalescent, rich in ferments and proteins, and poor in alkali. The trypsin in it may be secreted in an active form, and the secretion may be suspended by atropine and excited by pilocarpine. The chemical secretion is thin and watery, is less concentrated in ferments and protein, and is rich in alkali. The trypsin in it is secreted in an inactive form, and the secretion is not affected by the injection of atropine. The nervous secretion is small as compared with that due to secretin. The main factor involved in digestion is undoubtedly due to the chemical secretion.

Pavlov found that pancreatic juice from a fistula had little or no digestive action on proteins, but if brought into contact with the duodenal mucous membrane or an extract of the duodenal mucous membrane, it showed at once powerful proteolytic properties. This has been confirmed repeatedly. Evidently this proteolytic enzyme is secreted in a proenzyme form (trypsinogen) which is activated or converted to the active enzyme after it enters the intestine, possibly by something contained in the mucous membrane of the small intestine. Pavlov supposed this substance in the mucous membrane to be an enzyme, and since its action was upon another enzyme he called it a kinase, enterokinase, which by hydrolytic action upon trypsinogen converts it to active trypsin.

## DIGESTIVE ACTION OF PANCREATIC JUICE

The proteolytic action of pancreatic juice differs from that of gastric juice in that it takes place in a neutral, slightly acid, or markedly alkaline solution and the effect is more rapid, powerful, and more complete. Trypsin can change native protein that may have escaped the action of pepsin digestion (metaproteins, proteoses, peptones). The end products are amino acids, although much of the hydrolysis of peptones and especially peptids is accomplished by proteolytic enzymes in the wall of the intestine and in intestinal juice (erepsin).

In the normal digestion of proteins it is believed that they are broken down completely to the amino acids and are absorbed in that form, but it is possible that some absorption may take place of the simple peptid or polypeptid compounds. In any given digestive mixture the actual products formed depend on the length of time the enzymes are allowed to act, and on the conditions, favorable or unfavorable, under which they act.

The end products usually obtained most easily are tyrosine, leucine, aspartic acid, glutaminic acid, tryptophane, lysine, arginine, and histidine. The first two of these substances have been known for a long time and may be obtained easily in crystalline form from pancreatic digestions. If the enzymes are allowed to exert their complete action upon the protein, the end products are closely similar to those obtained by boiling protein with acids. The hydrolysis caused by the acids and enzymes seems to be nearly identical, although that caused by the acids is more complete and perhaps is attended by secondary reactions.

The numerous products obtained by this complete hydrolysis consist chiefly of amino acids, that is, organic acids containing one or more amino groups ($NH_2$) in direct union with carbon. Most of them are monoamino acids, that is, contain one $NH_2$ group. The nitrogen of the protein molecule appears in the split products chiefly as amino acids but also in small part as ammonia.
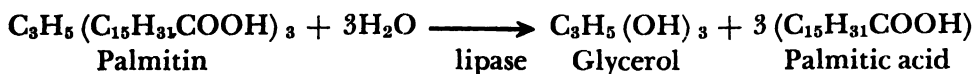
## ACTION OF THE DIASTATIC ENZYME (AMYLASE) OF THE PANCREATIC SECRETION

This enzyme may be readily demonstrated in the pancreatic secretion or in extracts of the gland. It causes a hydrolysis of starch with the final production of maltose. Maltose before absorption is acted upon by maltase of the intestinal secretion and converted to dextrose. Throughout the intestine, conditions are favorable for the conversion of starch to dextrose.

## ACTION OF THE LIPOLYTIC ENZYME (LIPASE, STEAPSIN)

It is now know that the pancreas secretes an enzyme capable of hydrolyzing or saponifying neutral fats. These latter bodies are chemically esters of

the trihydric alcohol glycerol. When hydrolyzed the fats break up into glycerol and their constituent fatty acids, in which form they are absorbed by the intestinal epithelium and again combined to form neutral fat characteristic of the body of the animal. The action of lipase may be represented, therefore, by the following reaction in the case of palmitin:

$$C_3H_5 (C_{15}H_{31}COOH)_3 + 3H_2O \xrightarrow{\text{lipase}} C_3H_5 (OH)_3 + 3 (C_{15}H_{31}COOH)$$
Palmitin            lipase    Glycerol        Palmitic acid

When lipase from any source is added to neutral oils, its splitting action is readily recognized by the development of an acid reaction due to the formation of the fatty acid. If a bit of fresh pancreas is added to butter, for example, and the mixture is kept at body temperature, the hydrolysis of fats is soon made evident by the rancid odor due to the butyric acid produced.

When pancreatic juice is mixed with oils or liquid fats, two phenomena are noticed: first, a splitting of the fat evidenced by an acid reaction due to the formation of fatty acids, and second, an emulsification of the fat. An oil is emulsified when it is broken up into minute globules that do not coalesce. The emulsification process is very striking. Artificial emulsions may be made by vigorous and prolonged shaking of the oil in a viscous solution of soap, mucilage, etc. Milk may be regarded as a natural emulsion that separates slowly on standing, as the fat rises to the top to form cream. When a little pancreatic juice is added to oil at body temperature, the mixture, after standing for some time, will emulsify readily with very little shaking, or even spontaneously. The emulsification is due to the formation of soaps. The lipase splits some of the fats, and the fatty acid liberated combines with the alakline salts present to form soaps. The presence of a small amount of soap formed in this way at the beginning of the reaction is then instrumental in causing the emulsification of the remainder of the fat. It was formerly believed that the formation of this fine permanent emulsion was the function of lipase and that these fine fat droplets were taken up directly by the villi epithelial cells. This view was strengthened by the histological fact that during the digestion of fats the epithelial cells may be shown to contain fine oil droplets in their interior. Recent work explains that the emulsification is a mechanical preparation of the fat for the hydrolysis to glycerin and fatty acids. Fat droplets observed in the epithelial cells during absorption are due to a resynthesis to the fat characteristic of the animal. The two products of the action of lipase, the glycerol and the fatty acids, are absorbed by the intestinal epithelium. The fatty acids themselves are insoluble in water, and it has been supposed that they form soaps with the sodium salts present and are absorbed in this form. Verzar and Kuthy (1929) emphasized the fact that alkali soaps are not stable in a solution with a pH below 9 and that the

intestinal contents during digestion have a neutral or even slightly acid reaction. They found that the fatty acids form soluble and diffusible compounds with the bile salts, sodium glycocholate, and sodium taurocholate which are stable at a pH of 6.2. They believe that fatty acids are absorbed in this form. After absorption, the two constituents are resynthesized to form neutral fats. In this synthesis the fatty acids are combined with glycerin in such proportions as largely to make the fat characteristic of the animal.

### INTESTINAL DIGESTION

In the intestines the food undergoes its most profound digestive changes, and here also the products of digestion are largely absorbed. As has already been pointed out, owing to the small size of the cavity of the proventriculus the food does not remain in this organ long. Therefore, the amount of gastric digestion taking place in this organ must be inconsequential. Also it has been pointed out that the relatively dry nature of the gizzard contents (average moisture content 44.2 per cent) makes it improbable that any considerable enzymatic action goes on in that organ. In other words, the secretion of the proventriculus probably does not make much, if any, digestive impression on the food until the duodenum is reached.

The duodenum extends from the gizzard to the place of entrance of the bile and pancreatic ducts. Its reaction is always acid, and it is probable that the gastric juice exerts much of the greater part of its action here.

The gastric juice of birds is similar in composition to that of mammals; pepsin, rennin, and hydrochloric acid being present. The peptic activity of the gastric juice of the goose is considerably weaker (one-tenth to one-twelfth) than that of the dog. Protein digestion with gastric juice is really a peptic hydrochloric acid digestion, the combination being essential. The end result is a conversion of more or less of the protein to simpler and more soluble forms (the molecules formed being from one-twelfth to one-fifteenth as large as the original). Such a digestion seems to be preliminary to a complete hydrolysis to the constituent amino acids in the intestine. The accepted view for many years has been that the protein molecules undergo hydrolysis in successive stages with the formation of smaller and more soluble molecules. The steps have been described as follows: native protein—acid metaprotein (syntonin) —primary proteoses—secondary proteoses—peptones. While there is no doubt that the end result of peptic digestion is the production of a soluble protein of smaller molecular weight than the original protein to which the name peptone is given, the interpretation of the process as a series of hydrolytic cleavages has been brought into question by more recent conceptions regarding the structure of the protein. As pepsin is apparently unable to resolve the molecule into its constituent amino acids as may be done by acids and some enzymes, it might then appear that pepsin may attack

some specific bond in the molecule or aggregate of molecules of which protein is composed.

## RENNIN (RENNET, CHYMOSIN OR CHYMASE)

Rennin is a coagulating enzyme and occurs as the zymogen prorennin, being activated by the hydrochloric acid. As far as our present knowledge goes, the action of rennin is confined to milk, changing the soluble protein casein into a solid form by two distinct steps. First, the rennin acts upon the casein of the milk, converting it to soluble paracasein. The paracasein then reacts with the calcium salts, forming the insoluble protein, calcium paracaseinate, which constitutes the curd or coagulum.

Lipase is said to be absent in the gastric juice of fish and birds.

The glands of Lieberkühn are present in the intestine. Brunner's glands are reported to be absent, however. Although no definite data are available, it is probably reasonable to suppose that the intestinal glands secrete an intestinal juice similar in composition to that of mammals, and that whereas the digestion in the first part of the intestine or duodenum is distinctly gastric in character, the intestinal digestion proceeds along with that of the gastric digestion but is greatly enhanced in the ileum as the pancreatic and bile ducts open in the intestine at a point which is taken to be the demarcation between the duodenum and ileum.

## ABSORPTION OF THE FOOD PRINCIPLES

Probably very little absorption occurs prior to the small intestine. Both the small and large intestines absorb readily and rapidly. The small intestine is the chief seat of absorption in carnivora and omnivora. It is also important in herbivora. The large intestine as an organ of absorption is of limited importance in carnivora and man.

The small intestine is studded with innumerable, barely macroscopic absorptive structures (villi) possessing both rich lymphatic and venous capillaries. A villus is composed of a projecting core of tunica propria covered with columnar epithelial cells lining the intestine. Near the axis of the villus is found a large lymph capillary known as a lacteal. It begins near the tip of the villus and enters a plexus of lymph vessels lying just on the inner side of the muscularis mucosa. At their origin the lacteals are often branched. The villus possesses a rich network of blood capillaries, and many lymphocytes are found in the meshes of its reticular stroma. Smooth muscle fibers derived from the muscularis mucosa enter the villus, to whose basement membrane many are attached. The villi increase the absorptive surface of the small intestine several times (man 7 to 18).

The villi of the duodenum in the chicken are the longest. The diameter of the small intestine diminishes from the duodenum to the rectum. In the

anterior portion of the ileum the villi are wider and shorter, even approaching a leaflike form in some places. Toward the posterior portion of the ileum the villi increase in length. There are well-developed lymphatic and blood capillary systems so that absorption takes place from these portions of the bowel.

The rate of digestion is relatively more rapid in birds than in mammals, especially in small birds. It is stated that the complete digestive process in small birds eating berries can take place in 10 minutes. A magpie may digest a mouse in 3 hours. Chickens require 12 to 14 hours for the digestion of grain. Vegetable foods are digested less rapidly than animal foods. Crude fiber is digested with difficulty. It is stated that the carbohydrate starch may be absorbed 100 per cent and albuminoid substances to the extent of 50 per cent. Vegetable fats are digested less rapidly.

The metabolism of birds is very marked. Small birds can stay alive for only a short time without food. It is estimated that from 10 to 20 per cent of the live weight is taken in as food daily. Birds have a rather high body temperature—from 40° to 43° C. (104–109.4° F.). Small birds have the highest temperature. The average for the chicken is 40.8° C. (105.2° F.), and for the pigeon, 41.8° C. (107.2° F.). While sleeping and especially while setting, the body temperature drops and may be 2° less than the usual temperature. Birds are in almost constant motion when not sleeping or setting. During productive activity, birds perform very intensive work. The total weight of eggs that are laid in a certain period may surpass the weight of the body several times. During the time of laying, the calcium content in the blood is more than doubled. When a bird seeks food, it seems to be guided only by its organs of vision; it does not seem to be able to pick up food in the dark.

### REFERENCES

Bayliss, W. M., and Starling, E. H.: 1902. The mechanism of pancreatic secretion. Jour. Physiol. 28:325.

Browne, T. G.: 1922. Some observations on the digestive system of the fowl. Jour. Comp. Path. and Therap. 35:12.

Calhoun, M. L.: 1933. The microscopic anatomy of the digestive tract of Gallus domesticus. Ia. St. Coll. Jour. Sci. 7:261.

Leasure, E. E., and Link, R. P.: 1940. Studies on the saliva of the hen. Poultry Sci. 19:131.

Vermeulen, H. A.: 1929. Anatomie und Physiologie des Geflügels. Handbuch der Geflügelkrankheiten und der Geflügelzucht, T. van Heelsbergen. Ferdinand Enke, Stuttgart.

Verzar, F., and Kuthy, A.: 1929. Die Bedeutung der Gallensäuren für die Fettresorption. Biochem. Zeitschr. 205:369.

# POULTRY GENETICS AS RELATED TO PATHOLOGY

By Nelson F. Waters, *United States Regional Poultry Research Laboratory, East Lansing, Michigan*

and

James H. Bywaters, *Research Poultryman,*
*Virginia Agricultural Experiment Station, Blacksburg, Virginia*

☆    ☆    ☆

To the breeder of all types of livestock and to the student of veterinary medicine whose life work is dedicated to the prevention and control of disease in animals, may it be stated that a better understanding of the laws of genetics will through application prevent the perpetuation of many of the anomalies now known to exist in animals.—*The authors.*

During the past two decades a better understanding and application of genetic principles has resulted in a marked improvement of poultry. It is probable that most of the desirable genetic characters exist in the more popular breeds of poultry. Unfortunately, a majority of these desirable genetic characters, though present in the breed as a group, are rarely, if ever, in the right combination. The problem faced by the poultry breeder is to so mate his birds that more of the desirable characters will be present in favorable combination in each of a large number of birds within the same breed. The poultry breeder must keep constantly in mind that there are many deleterious or unfavorable characters present in all breeds and strains of poultry. Genetic improvement consists not only in perpetuating the desirable characters but also in eliminating the undesirable ones.

## THE IMPORTANCE OF HEREDITY

A knowledge of the basic laws of inheritance should be understood by every poultry breeder who would attempt genetic improvement of poultry. The science of genetics has demonstrated beyond a doubt that the fundamental aspects of heredity are relatively simple. This statement may appear to be somewhat of an exaggeration to the beginning student of genetics. Long before the mechanism of inheritance was known, animal and plant breeders realized and made use of the partially correct statement that "like begets like." While it is a general truth that plants and animals do reproduce their kind in a broad sense, a very superficial examination will show that seldom, if ever, do we get two individuals that are identically alike, or even nearly so.

The science of genetics as such is still in its infancy; nevertheless, it is

probable that the basic and fundamental laws of inheritance are known, and no startling discoveries are apt to occur in the future.

A paper by Gregor Johann Mendel (1865) provided the first indication that genetics was an exact science and governed by certain laws of inheritance. It is probable that even Mendel did not fully realize the importance of his discovery. Certainly his researches and interpretations were not understood by contemporary scientists for it was not until deVries (1900), Correns (1900), and von Tschermak (1900), each independently, rediscovered the basic laws of inheritance as stated by Mendel.

Subsequent to the findings of Mendel it is yet to be proven that the same basic laws of inheritance do not apply to all forms of plant and animal life or, in fact, to all living organisms. Mendel pointed out that certain characters of the garden pea are inherited on a unit basis. His work was the foundation for the science of genetics and our present-day knowledge of the physical basis of inheritance. It is safe to say that the mechanisms of inheritance would still be a mystery if it were not for the existence, in both the plant and animal kingdom, of easily differentiated allelic pairs of genes.

It is not the purpose of this chapter to provide the elementary background necessary to the understanding of the mechanisms of inheritance. It is necessary, however, to review a few generalizations on the physical basis of inheritance in order to understand the underlying scheme embodied in genetics. Several very excellent textbooks are available and should be carefully reviewed by the student who would increase his knowledge of the modern concept of genetics as it applies to poultry (Jull, 1940; Lush, 1945; Sinnott and Dunn, 1939; Snyder, 1940; Sturtevant and Beadle, 1939; Walter, 1938).

### THE RELATIONSHIP OF GENETICS TO PATHOLOGY

Genetics bears an important role in, or relation to, pathology. Because of the very nature of pathology, by far the greatest proportion of specimens subjected to diagnosis are abnormal or defective in one or more respects. A few of these abnormalities or defects are genetic in origin. Many more are the direct result of a lack of genetic resistance to a specific pathogen. Genetic resistance or immunity to disease in both plants and animals is universally accepted as an established fact. It may be advanced on a factual basis that absence of a disease does not always imply innate immunity. Nevertheless, there is enough evidence accumulated in both plants and animals to demonstrate the importance of heredity in relation to disease.

The modern concept of disease recognizes the interdependence of three variables—the inherited capacity of the host, the innate characteristics of the disease-producing agent, or pathogen, and the environment which may materially influence the interaction of the host and the pathogen.

A student of veterinary medicine recognizes immediately that a wide knowledge of biological phenomena is necessary to a more accurate diagnosis on the animal with which he is working. The appearance of a diseased condition in one animal should lead to a closer inspection of the flock or herd as a whole. A lack of knowledge of either the epidemiology or cause of a disease may result in widespread dissemination not only through infection or malnutrition but also by defective germ plasm.

Both in human and animal pathology the student has been too prone to consider the individual host *per se* without due consideration of the genetic heritage of either the host or the pathogen, and the interrelation of the environment to both the host and the pathogen. There are numerous cases where pathological conditions exist which are directly caused by specific genes. The ability of the diagnostician to recognize and properly classify hereditary diseases and defects will add much to genetic knowledge and greatly assist in the ultimate eradication of heretofore uncurable conditions.

Strong (1929) states: "The solution to the problem of cancer lies entirely in the future. It may also be said to lie in the dark. Such being the case, it is fallacy for anyone to ignore entirely any possible approach to the solution. We cannot ignore entirely the problem of transplantation merely by maintaining that the findings up to date have been misinterpreted by investigators not trained in the science of genetics. . . . The use of the science of genetics in such a biological problem is not a vain approach." Later, Strong (1940) writes: "The directional path taken by the individual in the induction of specific tumor types is the resultant of the genetic constitution of the individual and is determined by heredity."

Little (1941a, 1941b) provides a resumé and bibliography on the genetics of spontaneous tumor formation and of tumor transplantation. Ample evidence is provided in his work to show that " . . . there is compelling evidence that the genetic constitution of an organism plays a part in determining whether or not it will develop a tumor or tumors."

A study of pathology involves structural and functional changes caused by disease. Before such pathological changes take place, a normal condition is assumed. A clear understanding of the normal morphology and physiology of an animal or its parts must be known by the student before the pathological picture is clear. In much the same way, genetics is not concerned alone with defects and abnormalities of various kinds. For each genetically abnormal condition found in an animal or plant, the geneticist must consider the opposing normal allel. Morgan (1922) states this relationship quite precisely: "It is true that the student of Mendelian heredity does not often trouble himself about the nature of the character that he studies. He is concerned rather with its mode of inheritance. But the geneticist knows that opposed to each defect-producing element in the germ plasm there is a

normal partner of that element which we call its allelomorph. We cannot study the inheritance of one member of such a pair of genes without at the same time studying the other. Hence, whatever we learn about those hereditary elements that stand for defects, we learn just as much about the behavior of the normal partners of those elements. In a word, heredity is not confined to a study of the shuffling of those genes that produce abnormal forms, but is equally concerned with what is going on when normal genes are redistributed. This method of pitting one gene against the other furnishes the only kind of information relating to heredity about which we have precise knowledge."

## GENETIC PRINCIPLES IN DISEASE CONTROL

As early as the eighteenth century, plant breeders recognized that certain varieties of wheat were more resistant to disease than others. Subsequently, breeders found it was possible with selection to establish varieties of wheat more or less resistant to specific diseases. Biffen (1905) demonstrated the resistance in certain varieties of wheat to mycotic stem rust and thus laid the foundation for important advances in the knowledge of heredity in disease. Rapid advances have been made in the study of resistance to disease in higher plants, and many valuable resistant varieties have resulted. Progress in the same direction with animals is limited because of practical and intrinsic difficulties. The advantages offered by plants, such as self-fertilization, shorter generation time, and the availability of large numbers with limited facilities, are not at the disposal of workers in the animal field.

Innate resistance of domestic fowl to the *Salmonella pullorum* has been demonstrated by Roberts and Card (1926, 1935) and by Roberts *et al.* (1939). The facts presented by these authors suggest that the difference between resistant and susceptible chickens is due to an inherited differential in the number of lymphocytes at the time of greatest susceptibility to pullorum disease, which is immediately after hatching.

Genetic resistance to fowl typhoid in the chicken is reported by Lambert and Knox (1932). Four generations of selection for resistance to a standard dose of fowl typhoid bacteria resulted in a decided decrease in mortality in the selected population. These authors, discussing the complexities of a genetic approach, state: "A genetic analysis of disease resistance presents many difficulties that are not encountered in the study of other quantitative characters. Perhaps the greatest of these difficulties lies in the establishment of definite criteria of resistance. While mortality probably furnishes the best index, it, obviously, does not help one to determine the various sub-lethal degrees of infection which take place in the surviving population."

The research of Webster and Hodes (1939) demonstrates the necessity of working with host material of known resistance or susceptibility. Highly

inbred lines of mice, resistant and susceptible to a standard test dose of *B. enteritidis,* were used as test material. Using such inbred mice having similar genes, these authors were successful in obtaining standardized reaction in transmission experiments.

Russell (1941) in his introductory comments on the part the geneticist may play in providing more useful material for experimental medicine, states that " . . . any geneticist who samples the recent literature in such fields as physiology, bacteriology, pathology, cancer research, and experimental medicine in general is struck by three points. First, most of the workers who are still using animals of uncertain origin could profit by the use of inbred stock. Second, even when inbred animals are used, they are frequently not utilized to their full value. Third, owing to a lack of understanding of the consequences of inbreeding, erroneous conclusions are sometimes drawn from the results obtained with inbred material."

## ENVIRONMENTAL INFLUENCES

No hypothesis is tenable which seeks to separate characters or traits that are inherited and those which are not. The living organism consists of a multitude of highly differentiated cells, each group or in fact each cell of which has a very complex form and function which is clearly integrated with the whole. These cell bodies are capable of functioning with more or less unity within a wide range of environmental conditions.

The statement has been made that there is an inherent basis for every phenomenon of life. This is essentially true, but we also recognize that the environment, for the most part, is an inseparable attribute of all life's phenomena. There is one very marked difference between heredity and environment. The genetic make-up of an organism is fixed and unchangeable while the environment changes constantly. Definitions of heredity and environment are more or less academic and are valuable only as a basis for discussion. Lush (1945) expresses clearly the present-day thought on heredity and environment: "In the strictest sense of the word, the question of whether a characteristic is hereditary or environmental has no meaning. Every characteristic is both hereditary and environmental, since it is the end result of a long chain of interactions of the genes with each other, with the environment, and with the intermediate products at each stage of development. The genes cannot develop the characteristic unless they have the proper environment, and no amount of attention to the environment will cause the characteristic to develop unless the necessary genes are present. If either the genes or the environment are changed, the characteristic which results from their interactions may be changed."

The environment appears to have less influence on certain genetic characteristics than on others. For example, a White Leghorn fowl of the Single-

Comb variety will develop a single comb regardless of environmental conditions. However, the possibility of such a bird succumbing to fowl typhoid will depend not only on its innate resistance to the pathogen, but on many environmental factors affecting both the host and the pathogen.

Webster (1939) writes that "studies now under way indicate that the level of resistance which is inherited can be altered by many environmental factors, entirely aside from specific vaccines or sera. Not the least of these factors, for example, is 'diet'."

An appreciation of the various genetic traits and their mode of inheritance comes only after a better understanding of the influence of the environment on these genetic traits is obtained. Observation will demonstrate rather quickly that there are relatively few known characteristics in poultry that are inherited on a unit-factor basis. For the most part such characters are confined to morphological and plumage color variations, and apparently they are not influenced to any great extent by the environment. The great bulk of physiological differences in poultry, both quantitative and qualitative, such as hatchability of the egg, resistance to disease, egg production, body weight, and egg weight, provide a major challenge to genetic investigators. Nearly all, if not all, of the physiological characters are influenced to a large extent by the environment surrounding the bird. The rate of growth attained by an animal is influenced markedly by the kinds and amounts of food fed. But parasitism, both internal and external, may retard growth even though ideal nutritional conditions be maintained. These are but few of the known environmental conditions surrounding a bird which will definitely limit growth.

### GENETICS OF PATHOGENIC ORGANISMS

The genetics of pathogenic organisms leads us into an unchallenged and fertile field. Rivers (1939) states that "Nothing is known of genetics in viruses. Therefore, one wonders whether it is proper to speak of virus mutations. However, in spite of complete lack of knowledge of virus genetics . . . it has long been recognized that viruses may vary under natural conditions and that some variations can be deliberately brought about by experimental procedures."

Pathogenic organisms in general have morphologic and physiologic characteristics which have caused workers to classify them into species, types, and strains. Frequently, such classifications are based on the pathogenicity of these organisms with relation to the host. Genetic change is suggested in the pathogen in that they mutate, hybridize, and recombine. It is still an open question whether, after successive generations, the host shows increased or decreased resistance to a virulent pathogen, or whether the pathogen has lost its virulence through genetic change. Riker's (1926, 1940) work bears out

the indications that variations in the environment may change the virulence of the pathogen.

Stakman (1940) states: "The problem of the genetics of pathogenic organisms is essentially the problem of their variation. That they do vary and that the variation sometimes is extremely important is common knowledge. But how, how much, and why do they vary? What are the limits of variation, both with respect to the kind and magnitude of the modified characters and with respect to their duration? Is the variation temporary or permanent; is it due to the effect of environmental factors or to genic changes either induced by the environment or independent of it? Can pathogenic microorganisms adapt themselves to new environmental conditions merely by being subjected to them? Can pathogens change in virulence as a result of host influence? Can they increase the virulence as a result of successive passages through a given host? Is the change, if it occurs, quite temporary; is it in the nature of a *Dauermodifikation;* or is it permanent and heritable? Or are the apparent changes due merely to natural or conscious selection of strains from a mixed population; and do new strains arise as a result of mutation and hybridization, as in the higher plants?"

Reed (1940) has stated that during the past 15 to 18 years the literature has been flooded with evidences of variability in pathogenic bacteria. The work of Arkwright (1920) points out that one of the most frequently observed variations in bacteria is a change of smooth to rough form. Not only do changes take place in the structure of the colony but there is also a change in virulence. The new type Rough is quite stable and seldom is there a reversion from Rough to Smooth. Pathologically, this change is of great importance in that the new variant no longer has the properties of the original colony. Apparently the new form Rough represents a genetic change in the organism. Of greater significance is the fact that such changes in the pathogen present diverse immunological reactions in the host.

## FURTHER APPLICATIONS OF GENETICS FOR DISEASE CONTROL

Medical science is more and more recognizing the advantages of using known genetic material in the fight against disease. Through the use of inbred stock it has been demonstrated for many disease conditions that susceptibility and resistance are definitely inherited according to Mendelian principles.

There are numerous examples of the effect of inheritance of morphological and noninfectious pathological conditions in the animal kingdom. It is probably true that the practicing veterinarian knows more about the hereditary anomalies in livestock than the practicing physician knows about hereditary anomalies in humans. For the most part this is due to the greater care and consequently greater knowledge used in the selection of livestock.

It has been stated, and may be repeated with emphasis, that man uses greater caution in the selection of animals for breeding purposes than in the selection of his own mate. Few of the better breeders of livestock will use animals of unknown ancestry for breeding purposes.

Cuénot (1908) demonstrated a dominant lethal associated with yellow coat color in mice in which the animals designated as YY perish before birth and the yellow coat color is maintained only in the heterozygous (Yy) condition. The exact cause of death of the homozygous yellow individual is not known, but it provides an excellent example of how a single gene can disturb the life processes of an individual to the extent that development is inhibited close to the time of conception.

A lethal anemia of mice associated with white spotting is reported by Little (1915). Here we have evidence of a physiological defect which can be partly compensated for by injection of whole blood from normal animals (Gowen and Gay, 1932).

Crew (1923) reports a pathological defect in Dexter cattle termed achondroplasia which results in extremely short legs, together with generalized morphological disturbances throughout the body. This defect occurs with regularity in about one-quarter of the calves of the Dexter breed of cattle. It is interesting to note that such matings also produce the Kerry-type cattle having longer legs and narrower heads than the Dexter cattle. In the case of Dexter cattle we find the desirable type of short-legged, broad-headed individual in a heterozygous condition which consequently does not breed true.

Wriedt (1925) and Mohr (1926, 1929) report the occurrence of achondroplasia in cattle that is less extreme in its manifestation and is recessive in nature.

Lush et al. (1930) report the condition in goats wherein there is a failure of one or both testicles to descend into the scrotum. Hadley and Warwick (1927), McKenzie (1931), and McPhee and Buckley (1934) report similar instances of cryptorchidism in other livestock. This defect is inherited as a recessive and is sex-limited in that it is only manifested in the male.

There are numerous other defects in the larger domestic animals which have a genetic basis. Some of the more common lethal defects are listed by Hutt (1934), Eaton (1937), and Lerner (1944).

### PRACTICAL BREEDING CONSIDERATIONS IN POULTRY

It is the ultimate aim of a breeder to obtain and perpetuate animals or plants in a homozygous condition for desirable genetic traits or characters.

This means that an individual must receive identical contributions from each of its parents and that such parents are capable only of transmitting identically the same genes for the character studied. If the characters studied

differ only in one or two pairs of genes, which are easily differentiated, it is relatively easy to obtain homozygous material with which to work. Unfortunately, the majority of characters of great economic importance in both plants and animals are dependent on the interaction of many pairs of genes.

The mating together of unlike animals promotes heterozygosity and maintains variability. The great bulk of plants and animals are in an extremely heterozygous condition, and it is the goal of the breeder to reduce this heterozygosity and at the same time increase their productive value. The inability to recognize favorable combinations within a relatively large population forces the geneticist to resort to selection of seemingly the most desirable material. The value of selection alone, however, is limited unless combined with a system of mating, such as inbreeding, which reduces heterozygosity.

The inability of selection, under any circumstance, to fix a heterozygous condition, such as roan color in Shorthorn cattle, is an example familiar to most students of animal husbandry. The selection and mating of roan-colored cattle will result in progeny having a ration of $\frac{1}{2}$ roan, $\frac{1}{4}$ red, and $\frac{1}{4}$ white. No amount of selection or inbreeding will change this ratio if the parents show the roan color. Much the same condition results when Blue Andalusian fowl are mated together, resulting in a progeny ratio of $\frac{1}{2}$ blue, $\frac{1}{4}$ black, and $\frac{1}{4}$ blue-splashed white.

Considerable genetic variation is present in nearly all of the important quantitative and qualitative characters of poultry. The variability present in such characters permits a continuous selection aiming towards permanent improvement. Variation in morphological and physiological characters is one of the great phenomena of nature without which improvement by genetic selection would be impossible.

For the most part poultry raisers maintain a flock of birds for economic reasons. Whenever a poultryman makes different combinations of matings, it is his optimistic hope that the progeny of such matings will either maintain the high standards of production found in the parents or show some improvement. Unfortunately, relatively few poultry breeders maintain adequate performance records to permit a comparison of parent and progeny.

Selection of breeding stock is all too frequently based on the fallacy that any chicken is still a chicken and the reasoning that all chickens are alike. Both biologically and in dollars and cents, such a premise is wrong.

In general, the entire system of breeding chickens is without a clear objective, and each generation consists of a population inherently as mixed as the previous generation. It represents a mass selection of breeding stock which is absolutely opposed to the promotion of general uniformity. It will not be possible to provide the farmer with written specifications describing any desired character and expect him to make progress. In the first place the farmer has very little or no control of the breeding stock from which his

birds are derived, and in the second place the average farmer is not primarily interested in poultry.

The small flock owner is definitely concerned with all genetic traits, or characters, which affect the growth and productivity of the birds in his flock. Among the more important genetic characters of economic importance are hatchability, viability, growth, adult weight, feather development, sexual maturity, egg production, and egg weight. It should be emphasized repeatedly, however, that there are many environmental conditions which may and do affect a bird's genetic ability to give complete expression for a given character. It is equally true, however, that no amount of feed or care will change the innate productivity of a bird.

### HATCHABILITY

A most important treatise on the hatchability of chicken eggs as influenced by environment and heredity is presented by Landauer (1941). This article is accompanied by an extensive bibliography. Landauer states: "It is altogether probable that only a beginning has been made in unraveling the enormously complex train of events to the end result of which we refer with the one word 'hatchability.' This simplification of language probably has led to the common fallacy of assuming that we are dealing with a simple problem, genetically or otherwise."

Considerable evidence is available to show the widespread occurrence of deviations from normal or desirable structure in chicken embryos. In contrast to the placental relationship between the fetus and dam in mammals, avian embryonic development is completed outside the body of the parent. This fact permits a more complete examination of the avian embryo. Despite this advantage surprisingly few lethals of known genetic origin have been observed. This is not due to a lack of embryonic material inasmuch as industry statistics indicate that approximately 35 per cent of all eggs fail to hatch. It is recognized that a large portion of this failure to hatch may be due to infertility. On this point, however, we do not know the exact percentage of infertility, in that death may occur during the very early hours of development and hence is not properly classified. Slight acquaintance with the hatchery industry will immediately inform the observer that much embryonic death is due primarily to the unfavorable environment surrounding the egg prior to and during incubation.

### VIABILITY

The ability of a chicken to live and at the same time remain productive has received the increasing attention of poultry investigators over the past two decades. Conservatively estimated, at least 15 per cent to 20 per cent of all female chickens hatched in this country die from disease before they

complete a laying year. Each year the loss from diseases and parasites of poultry is approximately $100,000,000. This is a staggering total and such a condition would not be allowed to exist for long among other domestic animals. For years the poultryman has tried various medicinal remedies in an effort to reduce poultry mortality. Certainly, if these measures have been successful there is no evidence to support it in the ever increasing mortality rate. A few parasiticides have been developed in recent years which show considerable promise. There are also specific biologicals which have been found highly successful and should receive increased usage. However, such biologicals are limited as control measures to a few disorders while the great bulk of poultry diseases continues to kill indiscriminately.

Carefully applied breeding procedures hold promise of eventually reducing the mortality caused by disease. Roberts and Card (1926, 1935), and Roberts *et al.* (1939) show clear evidence that heredity is an important factor in resistance and susceptibility to pullorum disease. Lambert and Knox (1932) have demonstrated that selective breeding decreased perceptibly the mortality from fowl typhoid. Asmundson and Biely (1932), Biely *et al.* (1933), Patterson *et al.* (1932), Lee *et al.* (1937), Gildow *et al.* (1940), Bearse *et al.* (1939), Marble (1939), Hutt *et al.* (1941, 1944), Sturkie (1943), Bostian and Dearstyne (1944), Taylor *et al.* (1943), Waters (1945, 1946), Waters and Prickett (1946), and others indicate that the genetic approach to controlling poultry diseases is encouraging.

From a strictly genetic consideration, uniformity for most any character which includes viability will be difficult to attain if birds from new sources are introduced into the breeding pens. Aside from complicating the breeding problem, introduction of unknown stock into a flock provides an excellent opportunity to bring in infectious and contagious diseases. One practical suggestion to the small flock owner would be to purchase replacements repeatedly from the same breeder (either directly or through the hatcheryman) whose birds live and produce well (Waters, 1944a, b, c, d, e). The small flock owner frequently holds to the old and misleading belief that "new blood" must be introduced frequently to obtain the best results. Many of the most successful breeders in the United States have maintained closed flocks for years with no subsequent loss in either reproductive ability or general health.

### GROWTH AND BODY WEIGHT

The inheritance of growth and adult body weight is of great practical importance to the poultry raiser, inasmuch as a large part of the poultry income is derived from the sale of poultry meat. The problem becomes greatly involved, for not only must the bird attain its adult body weight during a relatively short period of time, but gains in weight must be made

with an economy of feed consumption. Further, the distribution of flesh on the body is also part of the problem. The breeds of the domestic fowl vary from the small Bantam weighing less than 2 pounds at adult weight to the large Brahma weighing 12 pounds or more. Even within a breed there is often a range of from 4 to 9 pounds. There has been less selection for high egg production in the heavier breeds such as the Brahmas, Jersey Giants, and Orpingtons, and in consequence, production is generally at a lower level in these breeds. The White Leghorn is one of the smaller weight birds and is frequently referred to as an egg-producing type. From the standpoint of the small flock owner who desires both meat and eggs, the American breeds, such as the Plymouth Rock, Rhode Island Red, New Hampshire, and Wyandotte, weighing from about 5 to 10 pounds, are most popular.

Research has demonstrated that the inheritance of body weight is extremely complex. Fortunately, it is possible to obtain some measure of expression of body weight of the individual bird. There are individual differences in the rate of growth of birds, and some grow faster than others during the early periods even though the ultimate adult weight may be essentially the same. Because of this variation in early growth it is possible through proper observation to select birds showing the more rapid growth for breeding purposes. Another established fact is that all breeds of the domestic fowl attain their full growth at approximately 10 months of age. Increases in weight may occur after this period, but such gains are due chiefly to accumulations of fat. Selection of breeding stock on the basis of mature body weight should consist of picking the largest birds within a breed. This type of selection, although slow, will assist in maintaining body weight within a flock.

### FEATHER DEVELOPMENT

The rate of feathering on young birds is extremely important to the commercial broiler producer. However, it is probable that a large share of the total number of broilers produced by the industry originate from small flocks where the production of broilers is secondary. Much is known about the genetics of slow and rapid feathering during the first 10 days after hatching. In any discussion of rate of feathering, a distinction must be drawn between feather growth during the first 10 days and feather growth during the subsequent 3 months. Warren (1925) has demonstrated a dominant sex-linked, slow-feathering factor which permits a segregation of males and females at hatching time. A recent work of Darrow (1941) suggests a strong tendency for day-old chicks having the greater number and length of secondary wing feathers to be better feathered at the broiler age. The closest association is between well-developed tails at 10 days and good back feathering at 6 weeks of age. McClary and Bearse (1941) report an

autosomal factor for slow feathering in White Leghorns. This slow feathering is expressed in the absence of all tail feathers and secondary wing feathers and slow growth of primaries and body feathers until the chicks are 4 to 6 weeks of age. After 12 weeks of age it is impossible to distinguish the slow-feathering chicks from rapid-feathering chicks. Sturkie (1941) reports a naked factor which is a dominant autosomal. However, this character differs from those previously reported in that the bird more or less retains this naked condition throughout life. Aside from the factors for feathering already mentioned, there would appear to be additional factors for rate of feathering, the inheritance of which is yet unexplained. It should be stated that modifications in the diet, as well as other environmental conditions, may affect feather growth. From the breeders' standpoint the late feathering birds should be either discarded or adequately identified for later discarding. There is not enough evidence as yet to show that the slow or rapid feathering evinced at hatching is entirely responsible for the rate of feathering at a later date.

## EGG PRODUCTION

The production of eggs represents approximately 60 per cent of the total income derived from chickens. Because of this fact much attention has been given to the genetic factors responsible for egg production. Aside from certain color and morphological differences, the earliest genetic work with chickens concerned the inheritance of egg production. Pearl and Surface (1909, and later) published numerous articles which stimulated considerable research by other workers, and these theories are reviewed by Jull (1940).

Any discussion of the inheritance of egg production must of necessity consider many other physiological characters which alone or in combination affect the bird's ability to lay eggs. The fact that almost any one of these physiological traits is in itself exceedingly complex from a genetic viewpoint emphasizes the difficulties involved in such a study.

Knox (1930a) has shown adequately that in general the earlier a bird starts to lay, the more eggs it will lay during the first laying year. In this connection, however, as shown by Jull (1923), Kempster (1926), Maw and Maw (1928), and Knox (1930b), birds hatched too early or too late may show lower egg production even though the age at first egg is relatively early.

The rate of production, which can be described only as the number of eggs laid in a given unit of time, is influenced greatly by many conditions, both environmental and genetic. For example, it has been observed that under similar management many birds cease to lay during the winter months. This cessation has been termed by some investigators as "winter pause." It has been observed that certain breeds lay very few, if any, eggs during the winter months despite the most favorable management surrounding them.

Crosses between such non-winter layers and relatively heavy producers have resulted in ultimate segregation of both types as well as an intermediate condition. From such results, genetic interpretations have been made which suggest that "winter pause" is dependent on specific genes.

Broodiness is another trait which affects the rate of egg production as well as total annual egg production. Goodale *et al.* (1920), Hays (1924), Punnett and Bailey (1920), and Hays and Sanborn (1926, 1939) have contributed much to a study of the inheritance of this character. This early work has resulted in accepted theory that two dominant pairs of genes which are complementary in action are responsible for the broody condition. Later work by Roberts and Card (1933) suggests that a sex-linked gene may be in part responsible for the inheritance of the broody trait. In general hens do not lay eggs during the broody period. Thus, total yearly egg production is reduced in the presence of this character. For the most part broodiness is less prevalent in Leghorns and the Mediterranean breeds than in the heavier breeds, although selective breeding has greatly reduced the incidence in many of these heavier breeds.

The rate of laying is further affected by the persistency and intensity of production, both of which are related and hard to define in terms of genetic factors. Persistency of production has been described by some workers as the ability to produce eggs over a relatively long period of time; this period is generally figured from age at first egg to the beginning of the first annual moult. High intensity of production, among other criteria, involves long cycles of unbroken production in a bird as contrasted to short cycles of production frequently broken by periods of nonproductivity. Examples of high intensity might be cited wherein a bird will lay upwards of 50 eggs without cessation.

From the foregoing statements it is certain that the inheritance of egg production is dependent upon many different pairs of genes. It would be exceedingly difficult for the average poultryman to analyze flock production records and attempt selection for each of the many factors involved. One general recommendation would be to maintain flock averages which would provide the owner with figures for comparison with flock averages where production is more accurately known and considered highly desirable. Flock replacements thus could be made with some hope of improving poor-producing flocks by acquiring birds from these better flocks.

### SEXUAL MATURITY

Considerable variation is known to exist in the age at which a bird lays its first egg or more commonly expressed as the age at sexual maturity. Several workers, Hays (1924, 1944), Warren (1934), and Waters (1934), have shown that many genetic factors are involved in the inheritance of this

character among which at least one dominant autosomal and a dominant sex-linked factor are most prominent. This dominant condition is of value to the poultry breeder in that a selection of early sexual maturing birds for breeding purposes will assist in perpetuating this character in the flock. There would appear to be a rather close association between the rate of body growth and sexual maturity in that a high percentage of total growth must be attained before a bird is able to start production. Any factor or combination of factors either genetic or environmental which retard the growth of a bird may well influence the age at which a bird begins to lay. However, this does not necessarily mean that a bird will commence to lay as soon as it attains a certain weight. Waters (1934) shows that genes for sexual maturity are to some extent independent of genes influencing adult body weight. It is well established that the earlier in life a bird commences to lay, the smaller may be the size of the bird at that time. Contrary, however, to the thought frequently expressed by poultrymen, the adult body weight of a bird is not affected by this early sexual maturity. Birds laying at an early age, other conditions being equal and with no measurable retardation, will attain their full growth at approximately ten months of age.

## EGG WEIGHT

Poultrymen as a group should recognize to a greater extent the importance of egg weight to the poultry industry. Every egg produced which weighs less than 2 ounces is arbitrarily discriminated against when placed on the larger markets. Various investigators have frequently raised the question as to whether egg weight is a dominant or recessive character. The answer probably lies somewhere between these two points of view with a multiplicity of other factors, both genetic and environmental. The most important question to the poultry industry is: How can egg weight be improved through the application of breeding methods? Insofar as the practical breeder is concerned, any breeding program must be easily applicable to his problem whether it is in egg weight, egg production, hatchability, or any other genetic trait. For some reason, certainly not a sound biological reason, the poultry industry has accepted the 56.7 gram, or 2-ounce egg as a standard by which an egg is judged to be large or small. If, unwittingly, the poultry industry has adopted a market standard of egg weight which excludes over half the egg population as being too small, it should be at least cognizant of this fact in order to better understand the genetic problem.

Jull (1940) presented a thorough review of the literature on egg weight, together with a list of references of the most important work up to that time, indicating that there are many complex problems involved in a study of the inheritance of egg weight.

Various investigators have shown that there is considerable variation in

egg weight with relation to sexual maturity and body weight. Nutrition, temperature, and other environmental differences also influence egg weight. It has been shown that for the first laying year the least amount of variation in average egg weight occurs between the eleventh and eighteenth months of age. Attempts to measure the egg weight of a given bird should take into consideration this age factor. It is a common observation that the first eggs laid by a pullet are frequently small. This fact has led to the erroneous conclusion that early sexual maturity retards permanently the size of the egg. Investigations have demonstrated that the weight of the egg increases with the weight of the bird up to approximately ten months of age. Indications are that when the body weight and age are considered, it is possible to predict with some accuracy the ultimate weight of eggs produced by birds that have not attained their full growth.

Waters (1941) found that it is advisable to select dams whose egg weight is 60 grams or more in order to produce progeny whose eggs have an average weight of 56.7 grams, or about 2 ounces per egg. Other things being equal, a careful selection of dams having an egg weight of 60 grams will result in an increased number of daughters whose egg weight will satisfactorily meet the trade requirements established for either hatching or market purposes. The influence of the sire on the egg weight of his progeny is not as evident as the influence of the dam on her progeny. Waters (1941) has failed to find any measure of the egg weight transmissibility of the sire when the progeny's egg weights, dam's egg weight, sister's egg weight, or in fact, the egg weight influence of any combination of ancestors for three generations are considered.

Hays (1941), in contrast to the above, presents material which indicates that the sire as well as the dam is influential in transmitting genes for egg weight to the progeny. Likewise, Hutt and Bozivich (1946) present convincing data indicating that the male contributes genes which do affect the egg weight of his progeny. Such conflicting interpretations suggest the complexity of a genetic study on egg weight. The recommendations to the poultry breeder would be to give consideration to both sire and dam in a breeding program aiming at the improvement of egg weight.

### APPLICATION OF POULTRY BREEDING PRINCIPLES

It has been pointed out that selection, when combined with a planned system of mating, can do much to promote uniformity, not only for such visible characters as comb, color, and type, but for the more elusive characters which are extremely hard to measure. Years of carefully selected and well-planned matings by the better breeders have resulted in a marked increase in the general breed uniformity of the nation's poultry. This improvement has resulted in a decided decrease in the number of birds which can only be
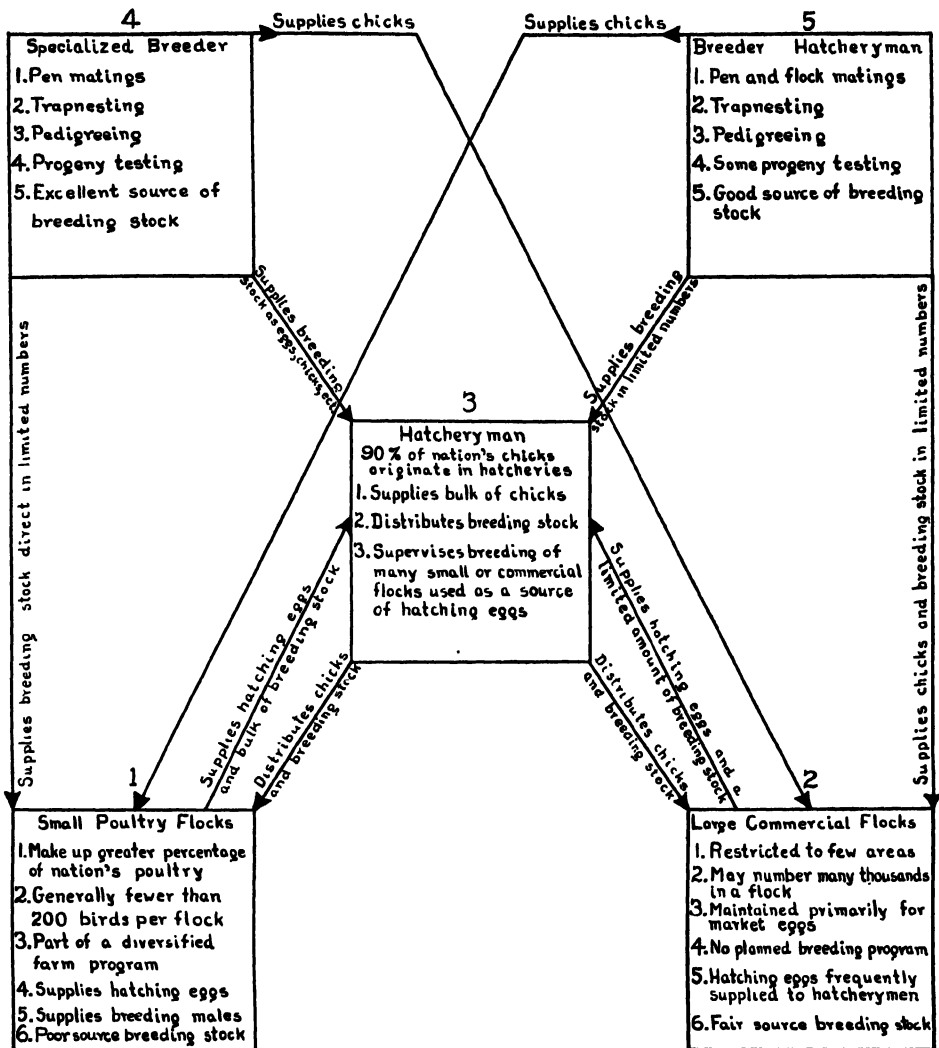
**4** — Supplies chicks → ← Supplies chicks — **5**

**Specialized Breeder**
1. Pen matings
2. Trapnesting
3. Pedigreeing
4. Progeny testing
5. Excellent source of breeding stock

**Breeder Hatcheryman**
1. Pen and flock matings
2. Trapnesting
3. Pedigreeing
4. Some progeny testing
5. Good source of breeding stock

Supplies breeding stock direct in limited numbers

Supplies breeding stock as eggs, chicks, etc.

Supplies breeding stock in limited numbers

Supplies chicks and breeding stock in limited numbers

**3**
**Hatchery man**
90% of nation's chicks originate in hatcheries
1. Supplies bulk of chicks
2. Distributes breeding stock
3. Supervises breeding of many small or commercial flocks used as a source of hatching eggs

Supplies hatching eggs and bulk of breeding stock

Distributes chicks and breeding stock

Supplies hatching eggs and limited amount of breeding stock

Distributes chicks and breeding stock

**1**
**Small Poultry Flocks**
1. Make up greater percentage of nation's poultry
2. Generally fewer than 200 birds per flock
3. Part of a diversified farm program
4. Supplies hatching eggs
5. Supplies breeding males
6. Poor source breeding stock

**2**
**Large Commercial Flocks**
1. Restricted to few areas
2. May number many thousands in a flock
3. Maintained primarily for market eggs
4. No planned breeding program
5. Hatching eggs frequently supplied to hatcherymen
6. Fair source breeding stock

FIG. 3.1. The interrelation of poultry enterprises in the United States.

classified as "barnyard fowl." Careful selection has resulted not only in the improvement of shape and color, but also for those economic characters which differentiate between a profitable and nonprofitable flock.

In the preceding discussion on practical breeding considerations, an attempt has been made to describe briefly what is known about some of the more important genetic characters in poultry.

A study of Figure 3.1 will assist in a better understanding of the breeding methods of the poultry industry. Any discussion of poultry breeding practices to be useful should take into consideration the different phases of the poultry

industry and the extent to which the breeding of poultry is involved. There are at least five easily defined types of poultry production enterprises, and the breeding activities of each of these influence to a large extent the quality of the nation's poultry. Every student of poultry husbandry should appreciate that the annual production of poultry and eggs in this country is valued at more than three billion dollars. Each year over one billion baby chicks are hatched and over one-half billion birds are raised to maturity. It is no easy task to recommend breeding practices which will include all phases of the industry and at the same time improve the quality of poultry and eggs. Nevertheless, in the past twenty years the quality of both poultry and eggs has improved, and annual production per bird has increased over this period. Some of this improvement can be attributed to better breeding practices, but the major share is probably due to improved management, housing, feeding, and sanitation.

Poultry enterprises may be classified and discussed as follows:

*1. Small flocks contributing a minor part of income.*

For the most part small poultry flocks make up, from the standpoint of numbers, the great bulk of the poultry population. In general these small flocks are maintained on the farm to supplement an income obtained through diversified farming. Very little, if any, planned breeding program is followed in such flocks. Replacements are obtained as baby chicks from hatcherymen who act as distributors. The farmer has very little or no control of the breeding stock from which these chicks are derived. Any improvement through breeding must come through the hatcherymen. It is true that many of these small flocks are used by the hatcherymen to provide hatching eggs for replacements. In such instances it is generally the task of the hatchery owner to supply breeding stock and to supervise breeding operations. Figure 3.2 shows that breeding stock for replacement flocks may come either direct or through the hatchery from specialized breeders. Unfortunately, the breeding stock used to propagate our poultry population, for the most part, is produced from parents which were selected only on the basis of appearance. Such selection over a period of years has improved the outward appearance of our poultry flocks, resulting in greater uniformity for breed and variety characteristics, but an accurate measure of productivity during this same period is difficult to obtain. It is true that scattered reports show an increase in average egg production for many sections of the country, but whether this improvement is due to better breeding methods, improved methods of culling, or to better management and feeding cannot be determined. It is becoming more evident that increased productivity of farm flocks will come through the poultry farmer's incessant demands on the breeder and hatchery distributors for chicks produced from the more productive parents of known breeding ability. The fact that the greatest buyer of baby chicks is the small poultry

flock owner means that the hatchery operators will listen when suggestions from these buyers are made. Every small flock owner should know something about the simple laws of inheritance and how they operate. They should know something about the standard requirements of egg size, body weight, viability, and breed characteristics. It is not amiss, however, to point out to the chick buyer that there are certain fundamental requirements of sanita-



High hatchability
Good viability
Large body weight
High egg production
Large egg size

BREEDING PROGRAM

1 Closed flock
2. Individual matings
   a. Trapnesting
   b Pedigreeing
   c. Progeny testing

High hatchability
Good viability
Large body weight
High egg production
Large egg size

A good start
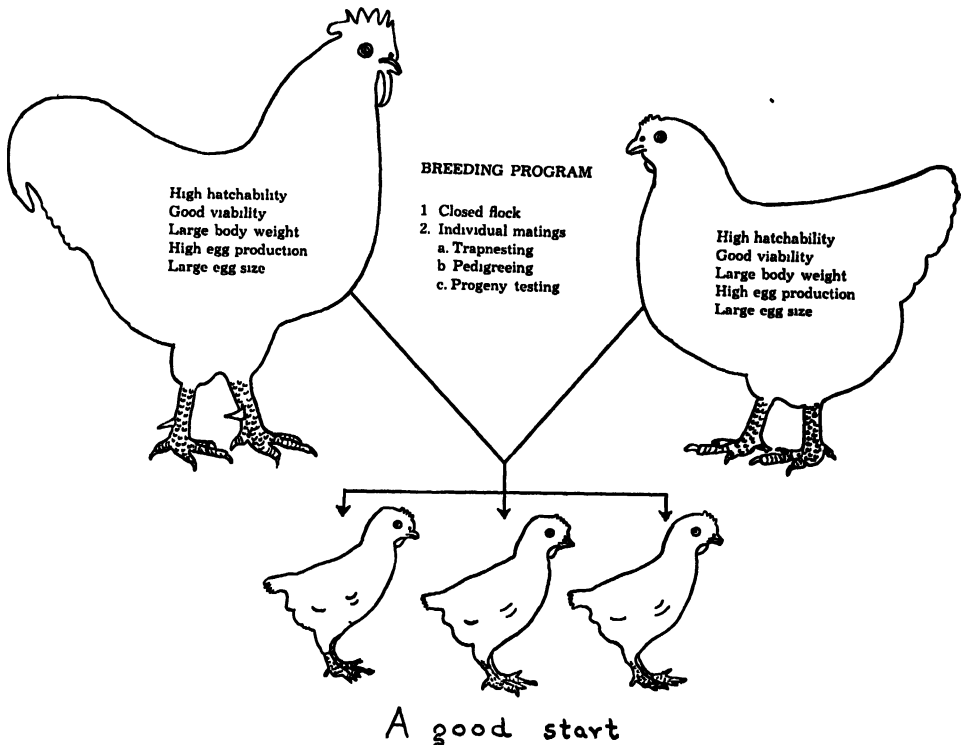
FIG. 3.2. Breeding birds from a closed, carefully selected flock will result in greater uniformity for those characters essential to a profitable flock.

tion, nutrition, and management without which no chick can grow to maximum productiveness.

*2. Large flocks contributing a major part of income.*

There are many large poultry flocks scattered throughout the country which provide either a major part of the farm income or the entire income. Such flocks are frequently termed commercial poultry flocks and may include many thousands of birds in a single flock. Similar to the small poultry flocks, little or no progressive breeding is followed in these larger flocks. Replacements are obtained for the most part from commercial hatcherymen. Occasionally the commercial flock owner obtains his replacements from specialized breeders who operate their own hatcheries. Such replacements, if

continued over a period of years, will tend toward greater flock uniformity from year to year. Not infrequently such flocks supply a large number of hatching eggs to the hatcheries. The owner of a large flock is often better informed and more critical of his source of supply than the small flock owner and may frequently consult with the hatchery owner regarding breeding procedures followed.

Success with the large commercial flocks is measured by the economic return of such flocks. From the economic standpoint this is sound business practice but too often it depends not on the breeding worth of the flock but on the ability of the owner to manage poultry and market poultry products.

### 3. The hatcheryman as a distributor of poultry.

The hatchery industry is responsible for the distribution of nearly 90 per cent of all the chicks produced in the United States. Every hatcheryman in the country is directly cooperating in a breeding program of stupendous size. No other phase of animal husbandry can possibly influence the breeding standards of the entire nation as quickly or efficiently as the hatchery industry. It would be of great value to study the poultry breeding practices of the hatcheryman and suggest procedures which will promote genetic improvement among our poultry population. A few years ago large hatchery operations now existing were unknown. Many flock owners practiced artificial incubation as a means of providing a larger number of chicks of uniform age. Other small flock owners would bring eggs to those owning incubators for custom hatching. The hatchery industry mushroomed to major importance with the introduction of larger and more efficient incubators and with better facilities for transporting baby chicks. The increased demand for chicks caused hatchery owners to seek poultry flocks that could supply hatching eggs in large quantities. Many problems of sanitation, disease, egg supply, distribution of chicks, and uniformity of product forced the hatcheryman to increase his knowledge of the poultry industry. The expansion of the hatchery industry has brought on new problems so fast that the poultry industry as a whole has had difficulty in keeping pace with developments. Besides the duties involved in managing the hatchery, the owner frequently tests for pullorum disease, culls undesirable birds, supervises breeding procedure, furnishes breeding stock, and in many cases acts as informant on management and nutrition.

The breeding stock producing the chicks distributed by the hatcheryman should come from the specialized breeder. Unfortunately, all too frequently the breeding stock producing the chicks distributed by the hatcheryman, is for the most part of unknown parentage. Breeding stock is obtained from distant areas, hatching eggs are obtained from many different sources representing unknown genetic potentialities, and chicks are distributed widely.

For the most part, if birds pass certain standard qualifications and resemble the breed, they are selected as potential breeders. One big problem of the poultry industry is to assist the hatcheryman to obtain stock of good ancestry at a cost that will not be prohibitive.

*4. Breeding flocks maintained by specialized breeders.*

The specialized poultry breeder devotes a major portion of his time to poultry improvement. It is this group of poultrymen who are responsible for much of the breed improvement witnessed in poultry during the past twenty years. The specialized poultry breeder must trapnest and pedigree all of his birds as a prerequisite for the selection of breeding stock. Records should be obtained on fertility, hatchability, viability, egg size, egg quality, egg production, sexual maturity, early and rapid feathering, growth rate, and body weight. In addition, breed and variety characteristics must be maintained. All of these records should be analyzed and serve as a basis for selection. This information may be likened to a set of carpenter tools, usable to most anyone but useless for careful work unless some element of skill or knowledge is present. It will be readily appreciated that the small flock owner or commercial flock owner has neither the time nor skill to make use of many of these records even though they were available. The improvement of breeding stock rightfully belongs in the hands of the skilled breeder and it is to this breeder we must turn for continued improvement of poultry flocks.

*5. Combination of specialized breeder and hatcheryman.*

The breeder-hatchery flock is fast assuming an important part in poultry breeding practices. Many hatcheries have found it impossible to obtain a sufficient and dependable quantity of desirable breeding stock. This has led to the establishment of breeding flocks by the hatcherymen, thus enabling them to control and regulate the source of breeding stock.

## GENERAL DISCUSSION

The preceding discussion of poultry enterprises demonstrates the complexity of the problem in attempting to recommend breeding practices which will be useful to all poultrymen. It becomes obvious immediately that any breeding program must seek the cooperation of the hatchery industry. The small flock owners are not primarily interested in poultry breeding, and it would be economically unsound to recommend that trapnesting and pedigreeing should be followed or that they follow a complicated breeding plan. Greater progress will come through educating the prospective buyer of baby chicks to a better understanding of the different methods of selecting breeding stock practiced by various hatcherymen. With this information the chick

buyer is in a position to make an intelligent choice. The hatcheryman must be provided with desirable breeding stock if genetic progress is to be expected. This breeding stock should come from the specialized breeder year after year, and this breeder in turn should maintain a closed, carefully selected flock. It is the specialized breeder who can follow best the principle of a closed flock. The maintenance of a closed breeding flock consists of closing the flock to the introduction of all outside breeding birds. A large number of the most successful breeders have not introduced outside breeding stock into their flocks for many years. Regardless of how well a group of birds may perform in one breeding flock, there is no guarantee that such birds used as breeding stock will improve or even maintain the standards of another flock.

After a hatcheryman is satisfied reasonably with the merits of the birds owned by a breeder, he should return to this same breeder each year for replacements. Each increase in the efficiency of the birds in the breeder's flock will result in an increase in the efficiency of the flocks using such birds as a breeding nucleus. The birds maintained by a breeder in a closed, carefully selected flock are approaching a degree of uniformity not attainable in those flocks where there has been a promiscuous introduction of questionable breeding birds year after year (see Figs. 3.2 and 3.3). As the specialized breeder's stock becomes more uniform, so will the hatcheryman purchasing such breeding stock disseminate more uniform birds to the small flock owner. The average hatcheryman is reluctant to return to the same source for breeding stock year after year. The belief is prevalent that "new blood" must be introduced frequently to obtain the best results. This is certainly a misconception on the part of any breeder or hatcheryman when large numbers of birds are involved. If only one or two males and several females were used, one might run into difficulties through inbreeding, but when 100 or more birds are used in a flock mating, there will be little, if any, danger incident to inbreeding.

### CROSSING OF BREEDS AND INBRED LINES

During the last four or five years there has arisen an increased interest in chickens resulting from the crossing of two or more pure breeds on the theory that such chickens will possess so-called "hybrid vigor."

The success of such crosses is dependent on many factors, among which are (1) the intrinsic quality of the parental stock, (2) the degree to which the parental stock complements each other in the offspring (how well they "nick" in terms of the animal husbandman), (3) the mode of inheritance of the characters not common to the parental breeds but which affect the market usefulness of the offspring or their products, and (4) the purpose for which the offspring are to be grown.

Thus, a cross between chickens of Breed I and chickens of Breed II may produce very desirable offspring while a cross between the same two breeds,

but from sources different from the first, may produce very undesirable offspring. The mere fact that a chicken is a crossbred between two or more specified (or unspecified) breeds is no assurance that it will be as good, or better, than a purebred chick for the purpose intended.

The same considerations apply to chickens produced by mating two or more inbred lines of the same or different breeds.

To the breeders who might be interested in selling such chicks, the best suggestion would be to try the crosses, on a small scale, and determine

? Hatchability
? Viability
? Body weight
? Egg production
? Egg size

NO BREEDING PROGRAM

? Hatchability
? Viability
? Body weight
? Egg production
? Egg size

A doubtful start
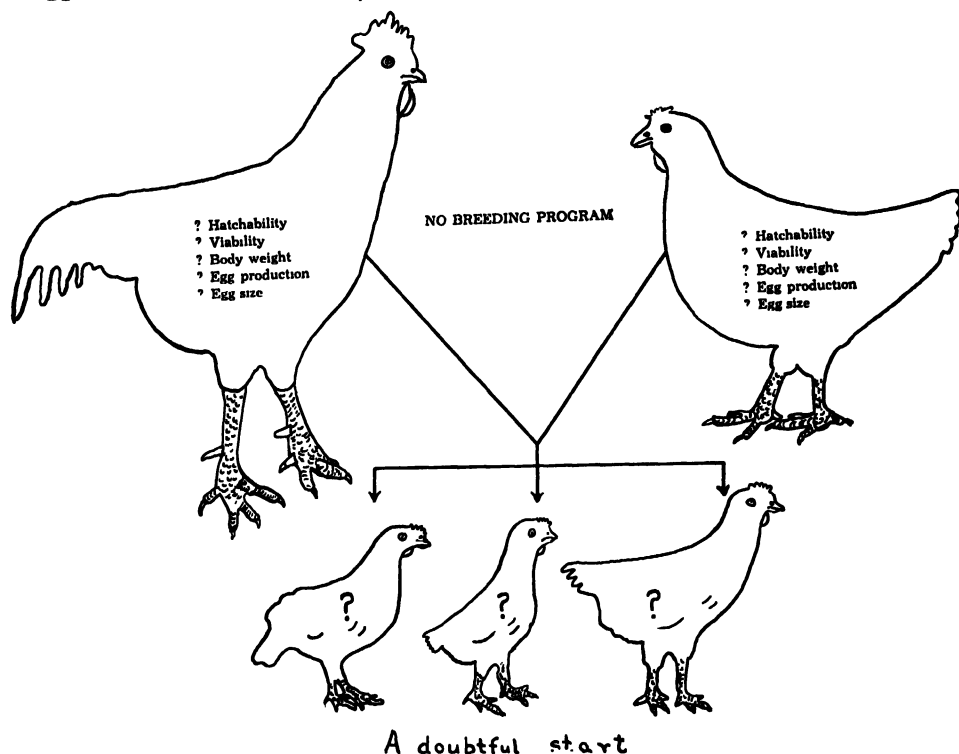
FIG. 3.3. The introduction of unknown breeding birds into a flock year after year will not promote uniformity in the offspring.

whether or not the offspring will improve or lower his reputation for producing high quality chicks. If he believes his reputation will be improved, he can expand his crossbreeding operations, staying as close to the original families as possible.

To the poultryman who buys chicks each year and who is interested in either type of crossbred chicks, it would be advisable to try a small number obtained from a breeder (probably through a hatchery) of good repute and compare their performance with that of the chicks he has been using. From such a trial he can decide whether he wants more from the same breeder next year.

## COOPERATIVE POULTRY PROGRAM

Within recent years it has become increasingly evident that one great need of the poultry industry is a cooperative undertaking between the flock owner, hatcheryman, and specialized breeder to bring about an improvement in the quality of hatching eggs, baby chicks, breeding stock, and market products. A realization of this need resulted in the inauguration of the National Poultry Improvement Plan (1935). The objectives of the Plan are "to improve the breeding and production qualities of poultry and to reduce losses from pullorum disease. This is being accomplished by: (1) The development of more effective State poultry improvement programs; (2) the identification of the quality of breeding stock, hatching eggs, and chicks by authorized terms that are uniform and applicable in all parts of the country; and (3) the establishment of an effective cooperative program through which newer knowledge and practical experience can be applied to the improvement of poultry and poultry products."

The Plan includes a breeding and selection program starting with the small flock owner and suggests progressive improvement, in cooperation with the hatchery industry, up to and including the highly specialized breeder. The Plan is a purely voluntary organization, and changes are made only by the participants. The accepted need of such a National Poultry Improvement Plan is witnessed by the fact that poultrymen in forty-seven states are now participating in some phase of the Plan. At the present time approximately one-half of all the chicks hatched commercially are produced under the Plan organization. This means that a large number of breeders and hatcherymen are making a conscientious effort to improve the quality of their chicks. Such continued organization and effort on the part of breeders and hatcherymen, combined with sound breeding procedures in their flocks, are bound to affect eventually the total breeding population of the country.

## REFERENCES

Arkwright, J. A.: 1920. Variation in bacteria in relation to agglutination by salts and by specific sera. Jour. Path. and Bact. 23:358.

Asmundson, V. S., and Biely, J.: 1932. Inheritance of resistance to fowl paralysis (*Neurolymphomatosis gallinarum*). I. Differences in susceptibility. Canad. Jour. Res. 6:171.

Bearse, G. E., McClary, C. F., and Miller, M. W.: 1939. The results of eight years' selection for disease resistance and susceptibility in White Leghorns. Poultry Sci. 18:400.

Biely, J., Palmer, V. E., Lerner, I. M., and Asmundson, V. S.: 1933. Inheritance of resistance to fowl paralysis, *Neurolymphomatosis gallinarum*. Science N. S. 78:42.

Biffen, R. H.: 1905. Mendel's laws of inheritance and wheat breeding. Jour. Agr. Sci., Cambridge 1:4.

Bostian, C. H., and Dearstyne, R. S.: 1944. The influence of breeding on the livability of poultry. N. C. Agr. Exper. Sta., Tech. Bul. 79.

Correns, C.: 1900. Gregor Mendel's Regel über das Verhalten der Nachkommenschaft der Rassenbastarde. Berliner Deutsch. Bot. Ges. 17:158

Crew, F. A. E.: 1923. The significance of an achondroplasia-like condition met with in cattle. Proc. Roy. Soc., Series B. 95:228-55.

Cuénot, L.: 1908. Sur quelques anomalies apparentes des proportions Mendéliennes. Arch. Zool. Exper. et Gén. 4e Serie 9:VII.

Darrow, M. I.: 1941. Relation of day-old chick wing feather development to feathering at the broiler age. Poultry Sci. 20:458.

de Vries, H.: 1900. Das Spaltungsgesetz der Bastarde. Berliner Deutsch. Bot. Ges. 18:83.

Eaton, O. N.: 1937. A summary of lethal characters in animals and man. Jour. Hered. 28:320.

Gildow, E. M., Williams, J. K., and Lampman, C. E.: 1940. The transmission of and resistance to fowl paralysis (lymphomatosis). Ida. Agr. Exper. Sta., Bul. 235.

Goodale, H D., Sanborn, R., and White, D.: 1920. Broodiness in domestic fowl. Data concerning its inheritance in the Rhode Island Red breed. Mass. Agr. Exper. Sta., Bul. 199.

Gowen, J. W., and Gay, E. H.: 1932. Physiological factors necessary to alleviate genetic lethal anemia in mice. Am. Nat. 66:289.

Hadley, F. B., and Warwick, B. L.: 1927. Inherited defects of livestock. Jour. Am. Vet. Med. Assn. 70:492.

Hays, F. A.: 1924. Inbreeding the Rhode Island Red fowl with special reference to winter egg production. Am. Nat. 58:43.

———: 1911. Transmitting ability in males of genes for egg size. Poultry Sci. 20:217.

———: 1944. The significance of inherited characters affecting egg production. Poultry Sci. 23:310.

——— and Sanborn, R.: 1926. Broodiness in relation to fecundity in the domestic fowl. Mass. Agr. Exper. Sta., Tech. Bul. 7.

——— and Sanborn, R.: 1939. Breeding for egg production. Mass. Agr. Exper. Sta., Bul. 307.

Hutt, F. B.: 1934. Inherited lethal characters in domestic animals. Cornell Vet. 24:1.

——— and Bozivich, H.: 1946. On the supposed matroclinous inheritance of egg size in the fowl. Poultry Sci. 25:554.

———, Cole, R. K., and Bruckner, J. H.: 1911. Four generations of fowls bred for resistance to neoplasma. Poultry Sci. 20:514.

———, Cole, R. K., Ball, M., Bruckner, J. H., and Ball, R. F.: 1941. A relation between environment to two weeks of age and mortality from lymphomatosis in adult fowls. Poultry Sci. 23:396.

Jull, M. A.: 1923. Early laying. Its economic significance. Agr. Gaz. of Canada 10:244.

———: 1940. Poultry Breeding. John Wiley and Sons, Inc., New York.

Kempster, H. L.: 1926. The relation of the date of sexual maturity to egg production. Mo. Agr. Exper. Sta., Res. Bul. 88.

Knox, C. W.: 1930a. Factors influencing egg production. II. The influence of the date of first egg upon maturity and egg production. Ia. Agr. Exper. Sta., Res. Bul. 128.

———: 1930b. Factors influencing egg production. III. The association of the date of hatch with date of first egg, sexual maturity and egg production in Single-Comb White Leghorns. Ia. Agr. Exper. Sta., Res. Bul. 152.

Lambert, W. V., and Knox, C. W.: 1932. Selection for resistance to fowl typhoid in the chicken with reference to its inheritance. Ia. Agr. Exper. Sta., Res. Bul. 153:261–95.

Landauer, W.: 1941. The hatchability of chicken eggs as influenced by environment and heredity. Storrs Agr. Exper. Sta., Bul. 216.

Lee, C. D., Wilcke, H. L., Murray, C., and Henderson, E. W.: 1937. Fowl leucosis. Jour. Am. Vet. Med. Assn. 91:146.

Lerner, I.: 1944. Lethal and sublethal characters in farm animals. A check-list and proposed numbering system. Jour. Hered. 35:219.

Little, C. C.: 1915. The inheritance of black-eyed white spotting in mice. Am. Nat. 49:727.

———: 1941a. The genetics of spontaneous tumor formation. In Biology of the Laboratory Mouse. C. D. Snell, editor. The Blakiston Co. Chap. 6:248.

———: 1941b. The genetics of tumor transplantation. Ibid. Chap. 7:279.

Lush, J. L.: 1945. Animal breeding plans. Iowa State College Press, Ames, Iowa.

———, Jones, J. M., and Dameron, W. H.: 1930. The inheritance of cryptorchidism in goats. Tex. Agr. Exper. Sta., Bul. 407.

McClary. C. F., and Bearse, G. E.: 1941. A recessive autosomal factor for slow feathering in Single-Comb White Leghorn chicks. Poultry Sci. 20:466.

McKenzie, F. F.: 1931. Anatomy of cryptorchid boars. Cryptorchidism in swine. Mo. Agr. Exper. Sta., Bul. 300:18.

McPhee. H. C., and Buckley, S. S.: 1934. Inheritance of cryptorchidism in swine. Jour. Hered. 25:295.

Marble. D. R.: 1939. Breeding poultry for viability. Pa. Agr. Exper. Sta., Bul. 377.

Maw, A. J. G., and Maw, W. A.: 1928. The variation in annual egg production according to the date laying commences. Sci. Agr. 9:201.

Mendel, Gregor Johann: 1865. Versuche über Pflanzen-Hybriden. Verhandl. Naturf. Verein, Brünn 4:3.

Mohr, O. L.: 1926. Über Letalfaktoren, mit Berücksichtigung ihres Verhaltens bei Haustieren und beim Menschen. Zeitschr. f. indukt. Abstam. u. Vererbungsl. 41:59.

———: 1929. Letalfaktoren bei Haustieren. Züchtungskunde 4:105.

Morgan, T. H.: 1922. Some possible bearings of genetics on pathology. (Middleton Goldsmith Lecture.) New Era Printing Co. Pp. 1–33.

National Poultry Improvement Plan: 1935. U. S. D. A., Animal Husb. Div. No. 14.

Patterson, F. D., Wilcke, H. L., Murray C., and Henderson, E. W.: 1932. So-called range paralysis of the chicken. Jour. Am. Vet. Med. Assn. 81:747

Pearl, R., and Surface, F. M.: 1909. Inheritance of fecundity. Me. Agr. Exper. Sta., Bul. 166.

Punnett, R. C., and Bailey, P. G.: 1920. Genetic studies in poultry. II. Inheritance of egg-colour and broodiness. Jour. Gen. 10:277.

Reed, G. B.: 1940. The genetics of pathogenic organisms. Problems in the variation of pathogenic bacteria. Occasional Pub. A.A.A. of Sci. 12:28–33.

Riker, A. J.: 1926. Studies on the influence of some environmental factors on the development of crown gall. Jour. Agr. Res. 32:83.

——: 1940. The genetics of pathogenic organisms. Bacteria pathogenic on plants. Occasional Pub. A.A.A. of Sci. 12:46–56.

Rivers, T. M.: 1939. Lane Medical Lectures. Viruses and virus diseases. Nature of viruses. Stan. Univ. Press, Calif. Chap. 4:77.

Roberts, E., and Card, L. E.: 1926. The inheritance of resistance to bacillary white diarrhea. Poultry Sci. 6:18.

—— and Card, L. E.: 1933. Inheritance of broodiness in the domestic fowl. Proc. Fifth World's Poultry Cong. 2:353.

—— and Card, L. E.: 1935. Inheritance of resistance to bacterial infection in animals. A genetic study of pullorum disease. Ill. Agr. Exper. Sta., Bul. 419:465–93.

——, Severns, J. M., and Card, L. E.: 1939. Nature of the hereditary factors for resistance and susceptibility to pullorum disease in the domestic fowl. Proc. Seventh World's Poultry Cong. 52.

Russell, W. L.: 1941. Inbred and hybrid animals and their value in research. In Biology of the Laboratory Mouse. C. D. Snell, editor. The Blakiston Co. Chap. 10:325.

Sinnott, E. W., and Dunn, L. C.: 1939. Principles of Genetics. McGraw-Hill Book Co., New York.

Snyder, L. H.: 1940. The Principles of Heredity. D. C. Heath and Co., Boston.

Stakman, E. C.: 1940. The genetics of pathogenic organisms. The need for research on the genetics of pathogenic organisms. Occasional Pub. A.A.A. of Sci. 12:9–17.

Strong, L. C.: 1929. Transplantation studies on tumors arising spontaneously in heterozygous individuals. Jour. Canc. Res. 13:103.

——: 1940. A genetic analysis of the induction of tumors by methylcholanthrene. Am. Jour. Canc. 39:347.

Sturkie, P. D.: 1941. A new type of nakedness in the domestic fowl. Poultry Sci. 20:474.

——: 1943. Five years of selection for viability in White Leghorn chickens. Poultry Sci. 22:155.

Sturtevant, A. H., and Beadle, G. W.: 1939. An Introduction to Genetics. W. B. Saunders Co., Philadelphia.

Taylor, L. W., Lerner, I. M., DeOme, K. B., and Beach, J. R.: 1943. Eight years of progeny-test selection for resistance and susceptibility to lymphomatosis. Poultry Sci. 22:339.

von Tschermak, E.: 1900. Über künstliche Kreuzung bei *Pisum sativum*. (Zeitschr. Landwirtsch. Versuchswesen in Oesterreich. 3:465.) Berliner Deutsch. Bot. Ges. 18:232.

Walter, H. E.: 1938. Genetics. The Macmillan Co., New York.

Warren, D. C.: 1925. Inheritance of rate of feathering in poultry. Jour. Hered. 16:13.

——: 1934. Inheritance of age at sexual maturity in the domestic fowl. Genetics 19:600.

Waters, N. F.: 1934. Growth and sexual maturity in Brahma and Leghorn fowl. Ia. St. Coll. Jour. of Sci. 8:367.

——: 1941. Genetic aspects of egg weight observed during inbreeding experiments. Poultry Sci. 20:14.

——: 1944a. Improving poultry through the closed flock system. U. S. Egg and Poultry Mag. 50:346–49, 375–76, 378–79.

——: 1944b. Close the flock to poultry diseases. U. S. Egg and Poultry Mag. 50:415–16, 424, 426–28.

——: 1944c. The hatcheryman's part in a closed flock system of breeding. U. S. Egg and Poultry Mag. 50:453–55, 472.

——: 1944d. The breeder's part in a closed flock system of breeding. U. S. Egg and Poultry Mag. 50:500.

——: 1944e. The flock owner's part in a closed flock system of breeeding. U. S. Egg and Poultry Mag. 50:549.

——: 1945. Breeding for resistance and susceptibility to avian lymphomatosis. Poultry Sci. 24:259.

——: 1946. The occurrence of lymphoid tumors in resistant and susceptible chickens. Jour. Hered. 37:281.

—— and Prickett, C. O.: 1946. Types of lymphomatosis among different inbred lines of chickens. Poultry Sci. 25:501.

Webster, L. T.: 1939. Inborn resistance to infectious disease. Sci. Month. 48:69.

—— and Hodes, H. L.: 1939. Role of inborn resistance factors in mouse populations infected with *Bacillus enteritidis*. Jour. Exper. Med. 70:193.

Wriedt, C.: 1925. Letale Faktoren. Zeitschr. f. Tierzüchtung und Züchtungsbiologie 3:223.

CHAPTER FOUR

# AVIAN HEMATOLOGY

*By* CARL OLSON, JR., *Department of Animal Pathology and Hygiene,
University of Nebraska, Lincoln, Nebraska*

☆　☆　☆

## INTRODUCTION

Hematology is defined as that branch of biology which treats of the
morphology of the blood and the blood-forming organs. When dealing with
the variations of the blood, it is essential that one consider not only the
cellular elements as they occur in the blood stream, but also the origin and
relationship of the blood cells and the relations between blood cells and the
cells of the connective tissues and the reticulo-endothelial system. Many
changes apparent in the peripheral blood are merely a manifestation of a
reaction taking place in the blood-forming tissues themselves. Such changes
should be studied at the site of primary disturbance in order to arrive at a
clear understanding of the process. There are some differences of opinion
concerning present-day conceptions of avian hematology and hematology in
its broader aspect. Some of these differences are due, in part, to the method
of study and technic employed by various investigators. There are some
phases of avian hematology upon which we do not have adequate informa-
tion, and in such instances we are inclined to find support of our knowledge
from work done on other forms of animal life. These analogies must be
drawn with extreme care.

The principal purpose of this section is to outline the salient points of
avian hematology. It will, therefore, be necessary to omit detailed discussion
of many debatable questions. Such discussions will be found in the references
listed at the end of the chapter. The domestic fowl or chicken is the main
subject considered, and except where noted all discussion refers to it.

### DESCRIPTIONS OF THE CELLS AND HEMOGLOBIN IN THE BLOOD

Numerous physiological factors influence the number of the various types
of cells and the amount of hemoglobin found in the blood. For this reason
it is not possible to give a single set of figures that may be regarded as fixed
normal values. The data in Table 1 are to be regarded as approximate values

for normal birds and should be used in connection with the knowledge available concerning physiological variations. The descriptions of the staining reactions of blood cells are of those secured by the use of Wright's blood stain or the May-Grünwald and Giemsa combinations of blood stains.

## Methods of Counting Blood Cells and Measuring Hemoglobin

Many procedures have been recommended for the enumeration of blood cells of birds. The fact that all blood cells of birds are nucleated precludes

TABLE 1

COUNTS OF ERYTHROCYTES AND VALUE FOR HEMOGLOBIN IN THE BLOOD OF BIRDS

| Bird | Sex | Erythro-cytes* | Hemo-globin† | Method of Measuring Hemoglobin | Observer |
|---|---|---|---|---|---|
| Chicken (Gallus domesticus)...... | Male | 3.23 | 11 76 | Photoelectric | Olson (1937) |
| | Female | 2.72 | 9.11 | " | "    " |
| Duck (Anas platy-rhynchos platy-rhynchos)........ | ...... | 3.06 | 15.6 | Photoelectric | Magath and Higgins (1934) |
| Pigeon (Columba domestica)....... | Male | 3.228 | 15.97 | Oxygen Capacity | Riddle and Braucher (1934) |
| | Female | 3.096 | 14.72 | "    " | "    "    "    " |
| Dove (Streptopelia risoria)......... | Male | 3.045 | 14.56 | "    " | "    "    "    " |
| | Female | 2.989 | 13.97 | "    " | "    "    "    " |
| Turkey........... | ... ... | ... .... | 10.7 | Newcomer | Dukes and Schwarte (1931) |
| Pheasant......... | ... | ... .... | 13.7 | " | "    "    "    " |
| Geese......... .. | ....... | ... .... | 14.9 | " | "    "    "    " |
| Swan........ ... | ....... | ... ... | 13.4 | " | "    "    "    " |
| Brants.......... | ..... .. | ... . | 14.7 | " | "    "    "    " |
| Peafowl......... | ..... . | ... . | 12.0 | " | "    "    "    " |
| Canary.......... | Female | 4.516 | 9.5 | Newcomer | Young (1937) |

\* Expressed in millions per mm.³
† Expressed in grams per 100 cc.

the use of the methods commonly employed for enumeration of mammalian blood cells. Relatively little difficulty is encountered in counting erythrocytes, but the counting of leukocytes introduces certain problems. The main objection to the available methods for counting leukocytes is the relatively large error associated with them. This error can be partially compensated for by making duplicate or triplicate counts and using the arithmetic average as representative of the true count.

The direct method of counting the leukocytes suspended in a suitable medium (as Toisson's fluid) in the hemocytometer is useful and satisfactory when one is dealing with normal blood. It is difficult to distinguish pathological immature red blood cells from leukocytes by this method. Under these conditions the direct method is obviously subject to error, and other methods are more suitable.

The method of Wiseman has been found to be fairly satisfactory for the routine study of chicken blood (Olson, 1935). The diluting fluid consists of 50 mg. of phloxine, 5 cc. of neutral formalin, and 95 cc. of Ringer's solution. The ordinary red blood cell diluting pipette is used, usually diluting the blood 200 times. A dilution of 1 to 100 will be advantageous in instances of marked anemia. The filled pipettes should be allowed to stand for several hours before the count is made in the hemocytometer. It is well to close the ends of the pipettes by stretching a heavy rubber band around the length of the pipette during this interval to avoid loss of fluid. The diluting fluid stains the erythrocytes a distinct pink color, and they may be counted in the usual manner; that is, the erythrocytes in 80 of the smallest squares are counted, and the result multiplied by 10,000, if the dilution of blood in the pipette is 1:200, will then represent the number of erythrocytes per cubic millimeter of blood. The count of leukocytes is obtained by counting the number of acidophilic granulocytes which are specifically stained by the dye phloxine in the entire ruled area of the hemocytometer (9 mm.$^2$). A differential count of leukocytes made from the stained blood smear provides the percentage value of acidophilic cells used in the calculation of the total number of leukocytes, according to the following formula:

$$\text{Total leukocyte count} = \frac{10}{9} \left[ \begin{array}{c} \text{Number of} \\ \text{acidophilic} \\ \text{cells*} \end{array} \right] \text{dilution} \left[ \frac{100}{\begin{array}{c} \text{percentage} \\ \text{acidophilic} \\ \text{cells**} \end{array}} \right]$$

A table of factors may be made taking into account the two variables and thus simplify the calculation to the multiplication of the factor corresponding to the percentage of acidophilic cells by the number of these cells found in the hemocytometer.

The number of thrombocytes may be estimated by counting the number of these cells which appear in the blood smear in conjunction with the 200 or more leukocytes observed in the process of making the differential leukocyte count. From the ratio thus found and the previously found total leukocyte count may be estimated the number of thrombocytes per cubic millimeter of blood.

The coefficients of variation or percentage error have been found in a series of counts of the blood cells to be approximately as follows (using phloxine diluting fluid) :

Counts of erythrocytes ........................ ........ 5.78 per cent
Counts of thrombocytes ............................. .23.66 "    "
Counts of total leukocytes ....................... .34.2  "    "

* Counted in hemocytometer.
** Found in blood smear.

FIG. 4.1.

The illustrated blood cells were from dry smear preparations stained with May-Grünwald and Giemsa combinations unless otherwise indicated.

1. Proerythroblast from marrow, transmissible granuloblastic leukosis. Wright's stain.

2. Lymphoid erythroblast from blood, transmissible erythroblastic leukosis. Wright's stain.

3. Lymphoid erythroblast from blood with leukemoid reaction, spontaneous lymphocytoma.

4. Polychrome erythroblast from blood, spontaneous erythroblastic leukosis.

5, 6, and 7. Polychrome erythrocytes from blood, spontaneous erythroblastic leukosis.

8, 9, 10, and 12. Erythrocytes from blood, normal.

11. Erythrocyte of blood from which nucleus has become lost.

13. Lymphoid stem cell or myeloblast from blood with leukemoid reaction, spontaneous lymphocytoma.

14 and 15. Lymphoid stem cells or myeloblasts from marrow, transmissible granuloblastic leukosis. Wright's stain.

16 and 18. Leukoblasts from blood with leukemoid reaction, spontaneous lymphocytoma.

17. Leukoblasts of Rieder type with lobulated nucleus from blood, transmissible granuloblastic leukosis. Wright's stain.

19 and 20. Metamyelocytes from blood with leukemoid reaction, spontaneous lymphocytoma.

21. Myelocyte from blood with leukemoid reaction, spontaneous lymphocytoma.

22. Myelocyte from blood, spontaneous erythroblastic leukosis.

23. Heterophil granulocyte with variation of granules from blood, spontaneous lymphocytoma.

24, 25, 26, and 27. Heterophil granulocytes, normal. Cell 27 shows swelling and rounding of granules as an artifact due to staining reaction.

28, 29, and 30. Eosinophilic granulocytes, normal.

31, 32, and 33. Basophilic granulocytes, normal. Cell 33 shows loss of water soluble basophilic material from granules.    ·

34, 35, 36, 37, 38, and 39. Lymphocytes, normal.

40 and 41. Thrombocytes, normal.

42. Mitotic figure in immature cell from blood with leukemoid reaction, spontaneous lymphocytoma.

43, 44, 45, and 46. Monocytes, normal.

47 to 54. *Plasmodium gallinaceum* parasites in erythrocytes of chickens. Blood films through courtesy of Dr. F. R. Beaudette, New Brunswick, N. J.

55 to 59. Haemoproteus parasites in erythrocytes of turkeys. Blood films through courtesy of Dr. F. R. Beaudette, New Brunswick, N. J.

60, 61, and 62. *Leucocytozoon smithi* and erythrocyte, blood of turkey. Blood films through courtesy of Dr. E. P. Johnson, Blacksburg, Va.

C. Olson.

10μ
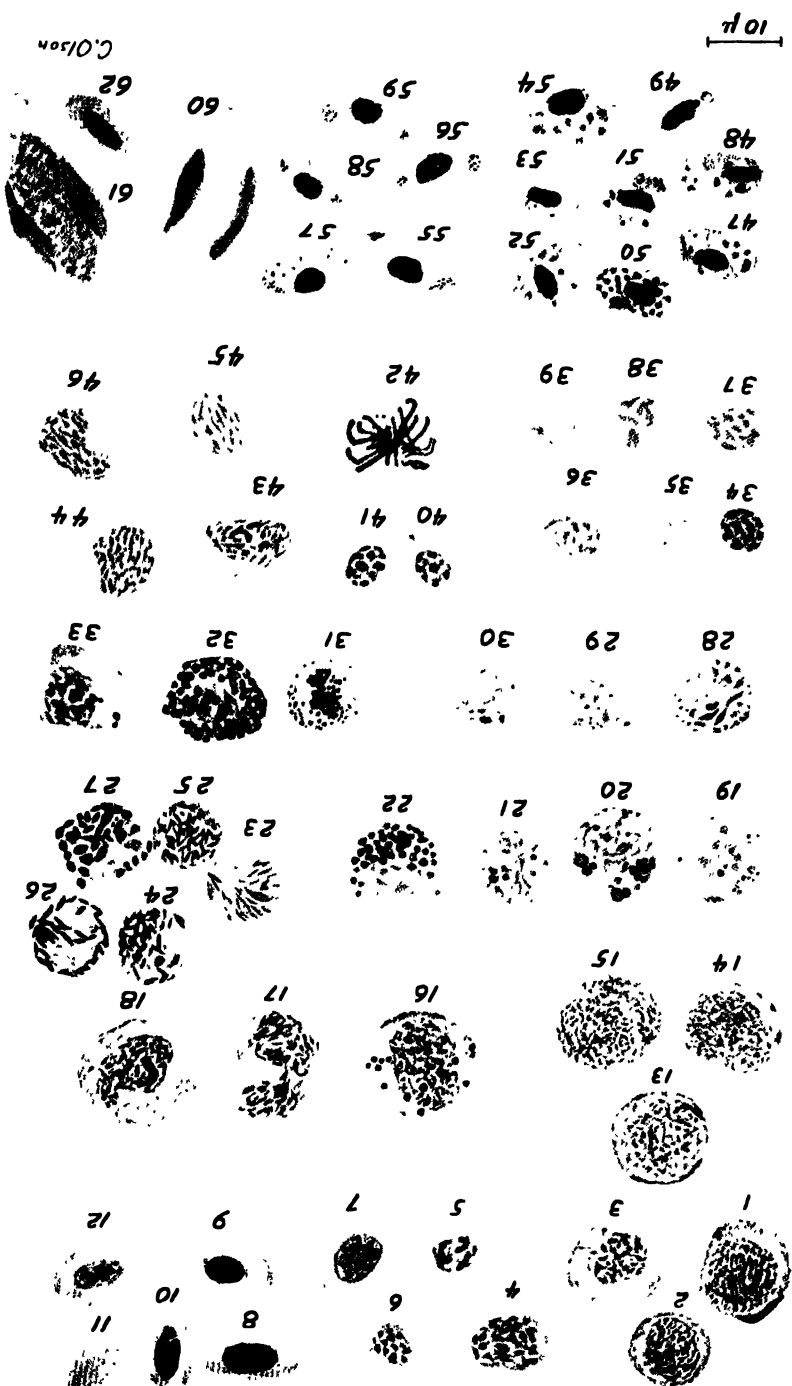
Differential leukocyte counts:

Lymphocytes ............................... 8.6  per cent
Heterophils  ................................27.96  "       "
Eosinophils  ................................58.84  "       "
Basophils  ................................62.68  "       "
Monocytes  ................................22.24  "       "

These figures indicate the order of accuracy that may be expected of a single count of blood cells. Theoretically, the error will be reduced by one-half if two complete counts are made and the average of the two used to represent the count. Such a procedure is to be recommended.

Differential counts of the leukocytes and morphological studies of the blood cells may be made from blood smear preparations. The basic staining elements (especially cell nuclei) of avian blood are much more numerous than those of mammalian blood; therefore, the technic usually employed for staining mammalian blood should be slightly altered in order to obtain the best results with avian blood. Different batches of staining solutions should always be tested by actual use and the staining time varied to suit the individual preparation. In the case of Wright's blood stain the length of time allowed for the stain to act may be varied. The staining time may also be varied with May-Grünwald stain. Giemsa stain may be varied both as to length of staining time and concentration. The May-Grünwald and Giemsa combination has been found useful especially with pathological blood as in leukosis when there are many basic staining elements. It is also invaluable for staining tissue imprint preparations. In such instances the concentration of the Giemsa solution should be increased.

The number of leukocytes may be expressed in terms of their relative number (percentage value as found in the differential count) or as an absolute number (actual number of cells per unit volume of blood, obtained by multiplying the total leukocyte count by the respective percentage value). The absolute count of the various types of leukocytes is a more reliable index to changes that may occur in an individual animal than the percentage value. A change may occur in the percentage value of one type of cell, due to an increase or decrease of another type of leukocyte, although the absolute value of the first type of cell remains the same. For example, given a total count of leukocytes of 20,000 cells and the differential count values of 25 per cent heterophils and 60 per cent lymphocytes, the absolute count would be 5,000 heterophils and 12,000 lymphocytes. Another differential count might be 12.5 per cent heterophils, 80 per cent lymphocytes, with a total count of 40,000 leukocytes, which at a glance, might be taken to indicate a decrease of heterophils by half and a slight increase of lymphocytes. Actually, the absolute values of the second count would be 5,000 heterophils,

or no change, and 32,000 lymphocytes, an increase of about two and a half times.

The hemoglobin may be readily measured by one of the acid hematin methods used for mammalian blood. It is necessary to correct the values thus obtained, as the nuclei of the red blood cells produce a turbidity causing the reading to be high. The hemoglobin readings obtained with a photoelectric hemoglobinometer measuring oxyhemoglobin concentration may be used without a correction factor. The readings of a Newcomer hemoglobinometer (measuring acid hematin concentration) may be converted into grams of hemoglobin per 100 cc. of blood as measured by a photoelectric hemoglobinometer by using the following equation:

Grams hemoglobin per 100 cc.$=0.16365$ Newcomer reading (percent)$-0.437$.
Slight differences occur in the color standards used in measuring acid hematin concentration, and precise comparisons between different instruments must be made with caution.

## Erythrocytes and Hemoglobin

The erythrocytes (red blood cells) of the bird are oval and nucleated. Variations in shape may be noted in the blood of a normal bird, and occasionally a spherical erythrocyte may be noted. The nuclei of the oval cells are likewise oval and of a mature character. They have relatively large irregular blocks of deep-staining chromatin material that is distinct from the more lightly staining parachromatin. The cytoplasm is orange in color. The round erythrocytes have a round, slightly less mature nucleus with smaller clumps of chromatin. Round erythrocytes without nuclei are occasionally seen. The average dimensions of chicken erythrocytes as found by Scarborough (1931–32) in his review of the literature were 12.2µ by 7.3µ. The red blood cells differ in size in different species of birds, and in general they are larger in the larger species. The principal function of the red blood cell is the transport of oxygen. This is done by means of the hemoglobin contained in the cell cytoplasm.

The count of erythrocytes and value for hemoglobin are usually higher in male birds than in female birds. This difference does not become apparent until about the time of sexual maturity. A hormonal influence is also indicated by the fact that juvenile and gonadectomized chickens of both sexes tend to have approximately the same counts of red blood cells. Domm and Taber (1946) have shown that androgens (testosterone proprionate) will cause an increase of erythrocytes in capons and five-month-old pullets. Younger pullets did not respond as well, indicating an age factor. Estrogen (alpha-estradiol benzoate) in large amount tended to counteract the effect of the androgen. Thyroidectomy of only male chickens caused a decrease of erythrocytes, whereas thiouracil treatment caused a decrease of erythrocytes

in males, females, and capons. Chicks at the time of hatching have a rela-- tively high level of hemoglobin, which soon drops to a fairly constant level. The number of erythrocytes and amount of hemoglobin vary with the season, the lowest values being found in the late summer and early fall, and the highest values in the winter (Olson, 1937). Domm and Taber (1946), making observation at three-month intervals, reported the lowest erythro- cyte values for hens in the winter and spring coinciding with egg production. The highest values were observed in the autumn. These workers noted no seasonal variation of red blood cells of males or capons. In pigeons and doves the males have higher values than the females; the lowest values for hemo- globin and erythrocytes are found in the summer; and the highest hemo- globin levels are noted in the winter, while the largest counts of erythrocytes are noted in the fall (Riddle and Braucher, 1934). A similar seasonal variation has been observed in canaries by Young (1937). A 48-hour starva- tion period was found by Palmer and Biely (1935b) to increase the counts of red blood cells. A diurnal variation of erythrocytes with high values at midnight and low values at noon has been observed (Domm and Taber, 1946). Cook and Harmon (1933) stated that the amount of hemoglobin varied with the intensity of egg production. They did not consider the effect of season, and others (see Olson, 1937) have questioned this statement. Diet will influence the level of hemoglobin, as iron sulfate or casein tend to increase the value (Cook and Harmon, 1933, and Cook, 1937). Hogan and Parrott (1940) found an anemia-preventing factor in the vitamin B complex which later was termed $B_c$ and found identical with folic acid (Odell and Hogan, 1943). Absence of this vitamin results in anemia, retardation of growth, and reduction of leukocytes and thrombocytes; a larger amount being necessary to maintain a normal level of leukocytes (Campbell, Brown, and Emmett, 1944). Harmon, Ogden, and Cook (1932) have demonstrated that the asphyxia of normal chickens will increase the value for hemoglobin from 5 to 25 per cent, whereas no increase is observed after asphyxia of splenecto- mized chickens. This demonstrates the reservoir function of the chicken's spleen. Pigeons subjected to sudden lowering of environmental air pressure for 8 hours had increased counts of erythrocytes and values for hemoglobin (Kocian, 1936). The factor of indoor versus outdoor environment was found to have no effect on the number of erythrocytes or level of hemoglobin (Olson, 1937).

*Polymorphonuclear Heterophilic Granulocytes (Heterophils)*

These cells are imperfectly round and have a diameter of approximately 10μ to 15μ. Their characteristic feature is the presence of many acidophilic crystalline granules in a clear colorless cytoplasm. In the chicken these granules are usually rod or spindle-shaped. Frequently, in routinely stained

smears, especially when Wright's stain is used, these granules are distorted in shape and appear as round or short rod forms. Magath and Higgins (1934) found the granules to be round in the heterophils of the duck. The nucleus is lobulated, with fairly heavy bands connecting the lobes. The number of lobes is usually two or three, and occasionally single or imperfectly lobulated nuclei may be observed. The chromatin and parachromatin arrangement is relatively heavy and coarse in the nucleus. The heterophils function in the defense mechanism against bacterial invasion as phagocytes that can be rapidly mobilized. These cells also have bactericidal action and the power to digest protein.

Male chickens have a slightly higher percentage of heterophils than do females, amounting to approximately 5 per cent. Heterophils also tend to be more numerous in the blood of older birds (Olson, 1937). Considerable variation in the percentage of these cells may occur with the passage of time in a given individual; this variation is somewhat less if one considers the absolute count only. Shaw (1933) reported a diurnal rhythm of the leukocytes of the pigeon. The relative count of lymphocytes is higher than the relative count of heterophils in the morning, but in the afternoon the relative count of heterophils may be greater, equal to, or slightly less than that of the lymphocytes. This is due to an increase in the absolute number of heterophils during the afternoon while the absolute count of lymphocytes remains constant.

*Polymorphonuclear Eosinophilic Granulocytes (Eosinophils)*

The eosinophils are of about the same size as the heterophils. They possess relatively large spherical granules whose color is dull red in contrast to those of the heterophil. Magath and Higgins (1934) found the granules of the duck's eosinophil to be rod-shaped. The cytoplasm has a faint bluish-gray tint. The nucleus is often bilobed, and the chromatin appears to be stained a richer blue than in the heterophil nucleus. The functions of eosinophils are not well understood. It is suspected that they act as a detoxifying power. In some animals and birds they are increased in verminous infestations and are found in the tissues in certain allergic states.

*Polymorphonuclear Basophilic Granulocytes (Basophils)*

The basophils are of about the same size and shape as the heterophils. The nucleus is usually masked by the mass of granules in the cells. It is weakly stained, and round or oval in shape, although often it is. lobulated. The cytoplasm is clear and colorless. Dark-staining, moderate-sized, basophilic granules are abundant. The material composing the granules is water soluble, and may be washed from the cell. More frequently the basophilic

material is incompletely washed, and the granules appear distorted and broken up. The function or functions of basophils or their tissue counterpart, the mast cells, are not known despite considerable investigation and many theories. Although there are relatively few basophils in the blood stream of the chicken, tissue mast cells are numerous. It is believed that the tissue mast cell may enter the blood stream and be identical with the basophil, and the reverse, that is, basophils leaving the circulation to become mast cells in the tissues, is considered likely in the chicken.

The number of basophils normally present in the blood is small. A slightly greater number may be found in the blood of young chickens than in the blood of adults.

*Lymphocytes*

The lymphocytes constitute the majority of leukocytes in the blood of the fowl. There is a wide range in the size and shape of these cells. In the past there has been a tendency to classify lymphocytes on the basis of size into large, medium, and small lymphocytes. Such a classification is entirely arbitrary as there are no sharply defined distinctions between such groups. The cytoplasm is usually weakly basophilic and may be confined to a narrow rim bordering one side of the nucleus, or it may constitute the major portion of the cell as in the case of the larger lymphocytes. The nucleus is usually round and may be slightly indented at one side. The chromatin pattern is usually rather coarse and blocky, especially in the small, more mature type of cells. In some instances the chromatin is rather fine and is not distinctly separated by the parachromatin material. Occasionally a few nonspecific azure granules may be noted in the cytoplasm especially near the point of indentation of the nucleus. It has been suggested with the support of some evidence that lymphocytes are capable of fixing toxic material and thus acting as a protective mechanism. Due to their high lipase content it has also been suggested that they participate in the digestion of fat. Studies in mammals have indicated that there are more lymphocytes entering the blood stream in 24 hours than are found in the circulation at any one time, which suggests a rather active circulation of these cells from the blood to the tissues and back to the blood again. Lymphocytes are abundant in the wall of the intestine, and many probably pass into the lumen and are lost. Lymphocytes in the tissues may differentiate into various types of cells. Mononuclear leukocytes or polyblasts are found forming a protective barrier between foci of chronic inflammation and healthy tissue. These cells are derived from the blood lymphocyte. It has been demonstrated that the resistance of mice to growth of inoculated cancer cells is directly related to the activity of lymphoid tissue.

The number of lymphocytes in the blood of adult female chickens is slightly greater than in the blood of adult male chickens. They are also

somewhat more numerous in the blood of young birds than that of adult chickens.

## Monocytes

The monocytes of the fowl are sometimes difficult to distinguish from the larger lymphocytes, and transitional forms between the two types of cells appear to exist in the blood. Generally, monocytes are large cells with relatively more cytoplasm than the large lymphocytes. The cytoplasm has a blue-gray tint. The nucleus is somewhat irregular in outline. The nuclear pattern is usually of a more delicate composition than in the lymphocyte, the chromatin having the tendency to appear in the form of strands rather than blocks. The functions of monocytes in the blood are not well understood. It is rather generally accepted that these cells may migrate into inflamed tissues and there hypertrophy to form large active phagocytes. They may then engulf not only bacteria but also particulate matter such as cellular debris; they also have the ability to digest such material. The monocyte is probably capable of differentiation into fibroblasts in the tissues.

Monocytes are more numerous in the blood of adult male chickens than that of adult female chickens. They are also more numerous in the blood of chickens kept in an outdoor environment than that of those confined indoors.

## Total Leukocyte Counts

The various factors influencing the number of specific types of leukocytes will obviously affect the total count of these cells. Some such physiological factors have been discussed above, and will not be considered here. The total number of leukocytes in the blood is greater in young chickens than in adult birds. Adult chickens kept in an outdoor environment have higher total leukocyte counts than those kept indoors (Olson, 1937). Palmer and Biely (1935d) reported that the counts of leukocytes tended to be high in birds kept in strict confinement. According to Palmer and Biely (1935a), the number of leukocytes in the blood of a chicken tends to fluctuate around a particular level characteristic of that individual. These workers (1935b) have also reported that there is an increase of leukocytes amounting to about 25 per cent, after a 48-hour fast. Hoppe (1935) noted a digestive leukocytosis with a maximum number of cells 4 to 5 hours after feeding, but only when the feeding period had been preceded by a 24-hour fast. Cook (1937) states that high counts of leukocytes are commonly found in chickens fed diets in which there is a lack of anti-hemorrhagic factor or minimal amounts of nitrogenous bases.

## Thrombocytes

These are the smallest cells seen in the blood of the fowl. They vary considerably in size and form. The typical thrombocyte is oval with a more

nearly round nucleus in the center of a clear cytoplasm. There are two or three small, brightly red staining granules at one pole of the cell. The chromatin of the nucleus is dense and is clumped into relatively coarse masses which are distinctly separated by the parachromatin. The thrombocytes are generally believed to play a part in the coagulation of the blood.

Thrombocytes are slightly more numerous in female than in male adult chickens, and also more numerous in young than in adult chickens.

The number of various types of cells and the amounts of hemoglobin in the blood of normal birds are listed in Tables 1 and 2, representing the values reported in the literature.

### THE ORIGIN OF BLOOD CELLS

In the embryo of the chicken the first blood cells are formed near the posterior portion of the germinal disc. These are found as blood islands in the wall of the vitelline sac from 21 to 24 hours after the beginning of incubation. These cells are mesoblastic in origin, have no hemoglobin, and are enclosed as small collections by what appears to be a lining endothelium. The

TABLE 2
COUNTS OF THROMBOCYTES AND LEUKOCYTES IN THE BLOOD OF BIRDS

| Bird | Sex | Throm-bocytes* | Leuko-cytes* | Differential Counts (Percentage) | | | | | Observer |
|---|---|---|---|---|---|---|---|---|---|
| | | | | Lym-pho-cytes | Het-ero-phils | Eo-sino-phils | Baso-phils | Mono-cytes | |
| Chicken (Gallus domesticus) .. | M | 25 4 | 19.8 | 59.1 | 27 2 | 1 9 | 1.7 | 10 2 | Olson (1937) |
| | F | 26 5 | 19 8 | 64 6 | 22.8 | 1 9 | 1 7 | 8 9 | "    " |
| Duck (Anas platyrhynchos platyrhynchos). | | 30.7 | 23 4 | 61 7 | 24.3 | 2 1 | 1 5 | 10 8 | Magath and Higgins (1934) |
| Pigeon A.M... | . | . | 13 05 | 65 6 | 23 | 2.2 | 2.6 | 6.6 | Shaw (1933) |
| P.M... | .. | | 18 55 | 47 8 | 42 8 | 1.9 | 2 4 | 5 1 | "    " |
| Turkey. . . | .. | .. | | 50 6 | 43 4 | 0 9 | 3 2 | 1 9 | Johnson and Lange (1939) |

* Expressed in thousands per mm.³

areas lined by endothelium become confluent, and a network of tubes develops. As development proceeds, the embryonal heart and vitelline veins are formed, which connect with the tubes in the area vasculosa to establish a circulation of the primitive blood elements. At about the end of the second day of incubation the circulation is established, and the primitive blood cells begin to acquire hemoglobin. They are still large round cells and multiply actively by mitosis. About the fourth or fifth day the hemoglobin-bearing

cells show a tendency to become elliptical in shape, thus assuming the definitive form of erythrocyte. At the time of formation of the definitive red blood cells there is still active multiplication of the primitive forms. Some of these are smaller and are very similar to the lymphoid germinal cells later developed in the connective tissues and hematopoietic organs. A few of these cells are the mother cells of the lymphocytes. At about the end of the second day of incubation, cells with round nuclei and basophilic cytoplasm appear in the tissues between the capillaries of the area vasculosa. These are the primordial lymphoid cells. Possibly some come from the blood islands at the time of formation of the endothelial lining, but for the most part they are derived from the mesenchymal tissue between the islands. At about the third or fourth day the primitive lymphoid cells develop round acidophilic granules in the cytoplasm. These granules are at first basophilic and soluble; later they become acidophilic, insoluble, and crystalloid in shape. The formation of blood in the wall of the vitelline sac becomes more active, reaching a maximum activity at the eleventh to twelfth day of incubation, and diminishing toward the eighteenth day. The blood cells of the young embryo are principally of the primitive type. These are gradually replaced by the definitive type cell. The replacement in the circulation of primitive erythroblasts by definitive types of red blood cells takes place at about the seventh day of incubation (Fennell, 1947). Small lymphocytes, granular leukocytes, and thrombocytes do not appear in the blood until the latter days of incubation. At the beginning of the twelfth day of incubation, the spleen begins to function as a hematopoietic organ. It produces lymphocytes, granular leukocytes, and erythrocytes at the expense of lymphoid cells very similar to the large lymphoid cells of the area vasculosa. The lympoid cells of the spleen are derived from the mesenchymal cells. Splenic hematopoiesis is especially active between the fourteenth and eighteenth days of incubation. The bone marrow is apparent and functioning at about the twelfth day. It is not highly active at this time, but gradually increases in activity until it is the principal source of blood cells at the time of hatching. A rapid rise in red blood cells and leukocytes was noted in the bone marrow from the time of hatching to the fifth day after hatching (Burmester, Severens, and Card, 1941). The liver is not an important hematopoietic organ in the chicken embryo. All of the mesenchyme distributed throughout the various parts of the body may form blood cells during embryonic life. Hematopoiesis is limited at first to the mesenchyme of the head, but beginning at about the fourth to-fifth day it is extended to the trunk, especially in the region of the aorta, and other parts of the body. Such activity is usually in the immediate vicinity of blood vessels. The mesenchyme may form not only lymphoid cells but granular leukocytes and erythrocytes as well. Toward the end of incubation this activity is restricted to certain organs as the bursa of Fabricius (where

erythrocytes may be formed up to the twenty-first day of incubation), thymus, spleen, bone marrow, wall of the intestine, and visceral connective tissue as in the pancreas and liver.

The formation of blood cells in the adult proceeds in a somewhat different manner from that outlined for the embryo. However, in times of stress upon the hematopoietic system or in some primary pathological conditions, there may be a partial return to the embryonic type of hematopoiesis. Erythrocytes and granulocytes are formed principally in the bone marrow. The erythrocytes are developed within sinuses lined by endothelial cells. Under normal conditions the young and immature erythrocytes are held within the sinuses during their development. When mature they are released to the circulation. Evidence indicates that the sinuses are in open communication with the blood circulation; however, the mechanism by which the immature cells are retained within the sinuses until maturity is not known. Granulocytes are formed principally in the bone marrow. Their locus of development is in the intersinusoidal areas and, therefore, extravascular. Under normal conditions they usually develop by multiplication of leukoblasts and pre-myelocytes, cell types already partially differentiated and fixed in their line of development. When the granulocytes have matured sufficiently they enter the circulation by migration into adjacent sinusoids. The myeloblast or hemocytoblast represents an ancestral cell type common to both granulocytes and erythrocytes. These cells occur in the marrow, and according to Jordan (1936) are represented by the germinal cells in the lymphoid nodules. His work indicates that the germinal cells produce small lymphocytes which surround them as a mantle. The lymphoid hemoblasts in the marrow may become either granulocytes or erythrocytes, depending on whether or not they gain access to the sinusoids before undergoing further development. The lymphoid hemoblasts or small lymphocytes may also form thromboblasts which multiply in the sinusoids and later become thrombocytes. Monocytes are formed from the small lymphocytes of the marrow. Jordan and Robeson (1942) believe the spleen is a normal source of the small lymphocytes in young pigeons. They demonstrated development of lymphoid foci in the marrow similar to those of the spleen as result of splenectomy. This increase of lymphoid tissue supplied the deficiency resulting from loss of the spleen.

The source of blood lymphocytes is chiefly the diffuse lymphoid tissue scattered along the intestinal tract, in the liver and in the spleen, together with the more or less organized nodular lymphoid tissue of the spleen, thymus, cecums, and palatine tonsils. Jordan (1936) believes that the small lymphocyte of the bone marrow (lymphoid hemoblast) is identical with that of the circulating blood. The circulating lymphocyte of the blood may then be regarded as a potential hemoblast. Likewise, the germinal center cells in the organized nodular lymphoid tissue are hemocytoblasts.

### THE RELATIONSHIPS OF BLOOD CELLS IN THE TISSUES

A consideration of the blood cells in the tissues leads to one of the most fascinating aspects of hematology, as well as to one of the most confusing. With the exception of the erythrocytes, all cells of the blood have important functions in the tissues outside of the blood stream, under normal as well as pathological conditions. Such cells as the heterophils fulfill their function in the tissues without change. The mononuclear leukocytes (lymphocytes and monocytes) are endowed with the ability of transformation into other types of cells in the tissues. Studies of such transformations have been made by various means. Morphology alone is a useful and important method of study, but it does not indicate function as well as does physiological study by means of tissue culture methods, supravital and intravital staining, and observations on the property of phagocytosis. The tissue cells of mammals have been studied more extensively than those of birds; however, it has been shown that analogous cell types exist in the chicken. This discussion is, therefore, rather general in nature.                                            .

Aschoff grouped an extensive and widespread system of phagocytic cells under the term reticulo-endothelium. The elements of this system are most numerous in the blood-forming organs. The cells belonging to this system have been given various names (fixed and free macrophages, clasmatocytes, Kupffer cells, active and resting wandering cells, histoblasts, littoral cells, histiocytes, adventitial cells, alveolar phagocytes of the lung, and others). Mann and Higgins (1938) suggest the use of the term histiocyte to designate the cells of this system. Thus there are the fixed histiocytes representing that portion of the system which lines the vascular sinusoids, such as the littoral cells of the liver, spleen, organized lymphoid tissue, and bone marrow. The free histiocytes include those elements of the reticulo-endothelial system found free in the tissues. The cells of the reticulo-endothelial system, the blood cells, and the fibroblastic cells have a common origin from the mesenchyme. Under many conditions they react as a group to protect the body from injury. The reticulo-endothelial system is in addition associated with the metabolism of iron, lipids, carbohydrates, and proteins, and the destruction of red blood cells as well as other processes of the body.

The free histiocytes of the tissues have many morphological variations. The term polyblast was suggested by Maximow as descriptive of the histiocytes observed in areas of inflammation. The free histiocytes are regarded as being derived principally from the lymphocytes and monocytes of the blood. Some may be formed by cell division of histiocytes and others by differentiation of fibroblastic elements. Some fixed histiocytes may become free and active. Transitional forms of histiocytes resembling fibroblasts are found in the tissues, suggesting the development of such cells into fibroblasts or the reverse. Both processes have been observed in tissue cultures. It should be

pointed out that tissue cultures of fibroblasts indicate the existence of different races and that the physiological properties and developmental tendency of such cells depend upon their origin and environment.

The fixed histiocytes are numerous in the sinusoids of the liver and spleen of the chicken where they actively phagocytize foreign particulate material. They exist in the lung as alveolar phagocytes, in the central nervous system as microglia, and in the loose connective tissue as sessile cells attached to the connective tissue fibrils. The fixed histiocytes may occur in any organ in the form of adventitial cells, in the adventitia of small blood vessels, and about the capillaries.

There is considerable variation in the amount of lymphoid tissue in the organs of normal chickens. The thymus and bursa of Fabricius increase rapidly in size after hatching of the chick and retrogress by the time of sexual maturity. The lymphoid tissue in the spleen, wall of the intestine, and periportal areas of the liver likewise varies in amount. Myeloid metaplasia is a term usually used to indicate the formation of granulocytes in organs other than the bone marrow. Such myeloid metaplasia may sometimes be observed in the lymphoid tissue of the thymus, periportal areas of the liver, and wall of the intestine. Huff and Bloom (1935) found marked extramedullary granulopoiesis and erythropoiesis in the spleen, liver, and kidney of canaries affected with malaria. The factors which control the amount of lymphoid tissue and degree of myeloid metaplasia are not well understood. In general it appears that age is one controlling element, as the lymphoid tissue is less active in older birds.

<center>THE BLOOD IN DISEASE</center>

Anemia is a condition in which the blood is deficient in either the number of erythrocytes or the amount of hemoglobin or both. The term is also sometimes applied when the erythrocytes show abnormal morphological features. Anemia may be associated with either acute or chronic disease, due either to suppression of blood cell production or to toxic destruction of blood cells. It may follow the loss of blood. Wirth and Kubasta (1939) have demonstrated the regenerative ability of the blood-forming tissues of the normal chicken by removing up to 85 per cent of the blood and observing the return of erythrocytes to normal levels within eight to nine days. The blood-forming organs of birds affected with disease will not respond with such rapidity. Anemia may result from the lack of constituents necessary for blood cell production, as in dietary deficiencies. The erythrocytes in anemia may show morphological deviations from the normal. The individual cells may be either larger or smaller than normal (anisocytosis). They may assume a variety of shapes as being pointed at one end, or round (poikilocytosis). Immature erythrocytes may be found in large numbers in the blood, depend-

ing upon the rate of production in the marrow. Many such immature cells are deficient in hemoglobin as indicated by the color of the cytoplasm which is blue-gray-yellow (polychromasia). It is important to distinguish between the blood picture of a severe regenerative anemia and that observed in erythroblastic leukosis. In the latter instance one may usually find the progenitors of the erythrocyte (lymphoid cells and lymphoid erythroblasts) in the blood. These are not usually present in the blood in simple anemia.

Leukocytosis is an increase in the number of leukocytes in the blood. Leukopenia is a decrease in number of leukocytes in the blood. As has been mentioned, there is considerable variation in the number of leukocytes in the blood of normal chickens. In many disease conditions, especially bacterial infections, there may be a leukocytosis. Various irritants may act in different ways. Some may attract a specific type of cell and repel others. Some toxic materials may stimulate the blood-forming tissues to production of nearly all types of leukocytes. Others may suppress the production of leukocytes and lead to a condition of leukopenia. Myelocytes containing preacidophilic granules that take a basic stain are described as occurring normally in the blood of the ostrich (Jackson, 1936). In marked heterophilic leukocytosis there may be myelocytes in the peripheral blood. Such a condition should not be mistaken for leukemia as it merely represents the tremendous effort on the part of the bone marrow to fulfill the demand for heterophils that do not have time to ripen before being pushed into the blood stream.

Anemia and leukocytosis may accompany many bacterial diseases. The study of the blood picture cannot be relied upon to serve as a differential diagnostic test in the group of infectious bacterial diseases. The increase should be regarded as an index of the response of the defense mechanism of the host against the infection. A study of the blood and blood-forming tissues is essential for the diagnosis and differentiation of leukosis from other diseases of the fowl. Infections with Salmonella species of bacteria provoke a marked heterophilic leukocytosis in chickens (Moore, 1895–96; Taylor, 1916; Cook and Dearstyne, 1934; and Olson and Goetchius, 1937). Anemia also is commonly present in such bacterial infections. Gauger, Greaves, and Cook (1940) found no difference between the counts of erythrocytes and leukocytes in the blood of pigeons that were positive to a serological test for paratyphoid infection and those that were negative. Ward (1904) has reported anemia and increased numbers of leukocytes in chickens with fowl cholera. Chickens spontaneously affected with tuberculosis have been found to have anemia and a leukocytosis of heterophils and monocytes (Olson and Feldman, 1936). Grimal (1938) studied the blood in two experimentally produced cases of the acute (Yersin) type of tuberculosis. A relative leukocytosis of lymphocytes was noted in the first week and of monocytes in the second, with a terminal relative leukocytosis of heterophils. These birds

died 23 and 25 days after inoculation. Pomeroy and Fenstermacher (1937) describe the findings in the blood of two turkeys with hemorrhagic enteritis. They observed anemia in both and a heterophilic leukocytosis in one.

Seastone (1935) observed a marked monocytosis in chickens with experimentally produced listerellosis. Following intravenous inoculation of the bacteria the monocytes increased from about 5,000 per mm.[3] to a peak of 60,000 per mm.[3] in five days, and at eight days dropped to about 10,000 per mm.[3] There was only a moderate increase of granulocytes and apparently no disturbance of the lymphocytes.

Thorp and Graham (1932) reported the results of counts of erythrocytes and leukocytes made on the blood of 71 chickens affected with acute laryngotracheitis. They found the number of erythrocytes to be within normal limits and the number of leukocytes to be slightly lower than the normal values for these cells as reported by other workers. It is pertinent to note that Thorp and Graham (1932) used the Blain method for counting leukocytes. This method has been observed to have a tendency to produce lower counts of leukocytes than other methods.

An increase of eosinophils has been noted in the blood of a chicken with coccidiosis and tapeworms (Yakimoff and Rastégäieff, 1929), in a grouse harboring *Trichostrongylos pergracilis* (Fantham, 1912), and in chickens with *Capillaria columbae* infection (Olson and Levine, 1939). A leukocytosis of heterophils and slight anemia were also noted in *Capillaria columbae* infection. Chickens with coccidiosis have been observed to have a relative heterophilic leukocytosis (Fantham, 1912) and an increase in the total number of leukocytes (Krömker, 1937). A controlled infection of chickens with *Heterakis gallinae* demonstrated an increase of heterophils and eosinophils, though the increase was not related to the number of parasites present (Wickware, 1947). Experimentally produced and natural cases of typhlohepatitis (*Histomonas meleagridis* infection) in turkeys were found by Johnson and Lange (1939) to be associated with a relative heterophilia and monocytosis. Except for a slight anemia of uncertain significance, no changes of the blood were observed by Olson (1935) in chickens before and after treatment for heavy infestations of lice. Yakimoff and Rastégäieff (1930) have noted a decrease in the percentage of lymphocytes and an increase in heterophils together with a slight relative decrease in monocytes in chickens experimentally afflicted with spirochaetosis. They also have observed a heterophilic leukocytosis in two spontaneous cases of spirochaetosis. Coles (1939) states that a marked anemia may occur in young chicks with aegyptianellosis, but that the infection in adult birds is associated with only a transient anemia. Jacobi (1939) found that the number of erythrocytes dropped to about one-third the normal level during the acute stage of experimental infection with *Plasmodium gallinaceum;* later the erythrocytes

and hemoglobin were slowly and simultaneously regenerated to their normal levels. Rostorfer and Rigdon (1946) have shown the anemia of ducks with experimental acute malaria to be of the macrocytic hypochromic type. There was also a decrease in functional hemoglobin in relation to the hemoglobin measured as acid hematin.

Krömker (1937) reported anemia to be associated with gout. He also studied the blood of a chicken with fowl pox and found an increase in the total number of leukocytes at the time the skin lesions were beginning to dry.

Poisoning from the ingestion of lead leads to anemia in wild ducks with basophilic stippling of the erythrocytes, according to the findings of Johns (1934). The question of whether or not basophilic stippling of erythrocytes occurs in lead intoxication in chickens is not settled. Keys (1924) cites Minot, and Meyer and Speroni to the effect that there is no stippling. Veenendaal (1935) reports having observed anemia without basophilic stippling in chickens with lead poisoning.

Lührs (1936) has described the changes in the blood of pigeons after exposure to high concentrations of war gases (phosgene and mustard). These changes were a relative increase in heterophils and a decrease in lymphocytes. The value for hemoglobin was low immediately preceding death of pigeons exposed to mustard gas. No characteristic degenerative changes of the blood cells were observed.

Under some circumstances of infection or disseminated neoplastic disease there may be a pouring out of unripe blood cells into the blood circulation. This leukemoid reaction may be so marked as to resemble a state of true leukosis. Sometimes there is sufficient evidence of granulomatous or metastatic neoplastic growth in the marrow to explain the reaction by a crowding out of the normal hematopoietic elements.

Marked changes occur in the bone marrow and peripheral blood in fowl leukosis. This disease and lymphocytoma are discussed in detail in another section, to which the reader is referred for a discussion of the hematology of these diseases.

The functions of the cells of the blood are of such a nature as to make them of great significance in both health and disease. Knowledge of the changes of the blood and blood-forming organs should constitute an important part of our information on diseases of birds. Such knowledge should consist of more than mere "counts" of the blood cells and embrace facts concerning the qualitative changes of cells such as indications of degeneration or regeneration of blood cells in the blood and tissues. Studies should be made at different stages of disease and the changes correlated with the course of the disease. Constant additions to our information are being made in the literature of today and should lead to better understanding of avian hematology in the future.

## REFERENCES

*Description of cells and hemoglobin in the blood:*

Biely, J., and Palmer, E. I.: 1935. *See* Palmer.
Bunting, C. H.: 1938. Functions of the leukocytes. In Downey, Hal: Handbook of Hematology. New York, Paul B. Hoeber, Inc., Vol. I, 438.
Campbell, C. J., Brown, R. A., and Emmett, A. D.: 1944. Influence of crystalline vitamin B₆ on hematopoiesis in the chick. Jour. Biol. Chem. 152:483.
Cook, S. F.: 1937. A study of the blood picture of poultry and its diagnostic significance. Poultry Sci. 16:291.
——— and Harmon, I. W.: 1933. The regulation of the hemoglobin level in poultry. Am. Jour. Physiol. 105:407.
Domm, L. V., and Taber, E.: 1946. Endocrine factors controlling erythrocyte concentration in the blood of the domestic fowl. Physiol. Zool. 19:258.
Dukes, H. H., and Schwarte, L. H.: 1931. The hemoglobin content of the blood of fowls. Am. Jour. Physiol. 96:89.
Harmon, I. W., Ogden, E., and Cook, S. F.: 1932. The reservoir function of the spleen in fowls. Am. Jour. Physiol. 100:99.
Hogan, A. G., and Parrott, E. M.: 1940. Anemia in chicks caused by a vitamin deficiency. Jour. Biol. Chem. 132:507.
Hoppe, R.: 1935. Beobachtungen über die Verdauungsleukocytose bei Hühnern, Wiadomóści Weterynaryjne 14:41, 1935. Abst. in Jahresb. Vet. Med. 58:210.
Johnson, E. P., and Lange, C. J.: 1939. Blood alterations in typhlohepatitis of turkeys, with notes on the disease. Jour. Parasit. 25:157.
Jordan, H. E.: 1938. Comparative hematology. In Downey, Hal: Handbook of Hematology. New York, Paul B. Hoeber, Inc., Vol. II, 700.
Kocian, V.: 1936. La composition morphologique du sang des oiseaux suivant les variations, de la pression atmosphérique. Soc. Biol. (Paris) Compt. Rend. 122:730.
Magath, T. B., and Higgins, G. M.: 1934. The blood of the normal duck. Folia Haematol. 51:230.
Oberling, Ch., and Guérin, M.: 1934. La leucémie érythroblastique ou érythroblastose transmissible des poules. Bul. de l'Assn. franc p. l'étude du cancer 23:38.
O'Dell, B. L., and Hogan, A. G.: 1943. Additional observations on the chick antianemia vitamin. Jour. Biol. Chem. 149:323.
Olson, C.: 1935. Available methods for examination of the blood of the fowl. Jour. Am. Vet. Med. Assn. 86:474.
———: 1937. Variations in the cells and hemoglobin content in the blood of the normal domestic chicken. Cornell Vet. 27:235.
Palmer, E. I., and Biely, J.: 1935a. Studies of total erythrocyte and leucocyte counts of fowls. I. Repeated erythrocyte and leucocyte counts. Folia Haematol. 53:143.
———: 1935b. II. Effect of 48-hour starvation on total erythrocyte and leukocyte counts. Jour. Am. Vet. Med. Assn. 86:594.
——— (Biely and Palmer) : 1935c. III. Variation in number of blood cells of normal fowl. Çanad. Jour. Res. Sect. D., Zool. Sci. 13:61.
———: 1935d. IV. Erythrocyte and leucocyte counts of birds raised in confinement. Canad. Jour. Res. Sect. D., Zool. Sci. 13:85.
Riddle, O., and Braucher, P. F.: 1934. Hemoglobin and erythrocyte differences according to sex and season in doves and pigeons. Am. Jour. Physiol. 108:554.
Scarborough, R. A.: 1931–32. The blood picture of normal laboratory animals. Yale Jour. Biol. and Med. 4: The chicken 202–6. Birds 323–24.
Shaw, A. F. B.: 1933. The leucocytes of the pigeon with special reference to a diurnal rhythm. Jour. Path. and Bacteriol. 37:411.
Young, M. D.: 1937. Erythrocyte counts and hemoglobin concentration in normal female canaries. Jour. Parasit. 23:424.


*Origins of blood cells and relationships of blood cells in the tissues:*

Bloom, W.: 1938. Fibroblasts and macrophages. In Downey, Hal: Handbook of Hematology. New York, Paul B. Hoeber, Inc., Vol. II, 1336–73.
———: 1938. Tissue cultures of blood and blood-forming tissues. In Downey, Hal: Handbook of Hematology. New York, Paul B. Hoeber, Inc., Vol. II, 1470–1585.
Burmester, B. R., Severens, J. M., and Roberts, E.: 1941. Blood cells in the bone marrow of the chick before and after hatching. Poultry Sci. 20:391.
Huff, C. G., and Bloom, W.: 1935. A malarial parasite infecting all blood and blood-forming cells of birds. Jour. Infect. Dis. 57:315.
Jaffé, R. H.: 1938. The reticulo-endothelial system. In Downey, Hal: Handbook of Hematology. New York, Paul B. Hoeber, Inc., Vol. II, 974–1271.

Jordan, H. E.: 1936. The relation of lymphoid tissue to the process of blood production in avian bone marrow. Am. Jour. Anat. 59:249.
—— and Johnson, E. P.: 1935. Erythrocyte production in the bone marrow of the pigeon. Am. Jour. Anat. 56:71.
—— and Robeson, J. M.: 1942. The production of lymphoid nodules in the bone marrow of the domestic pigeon, following splenectomy. Am. Jour. Anat. 71:181.
Jolly, J.: 1923. Traité Technique d'Hématologie. Paris, A. Maloine et Fils. Vols. I and II, p. 1131.
Mann, F. C., and Higgins, G. M.: 1938. The system of fixed histiocytes in the liver. In Downey, Hal: Handbook of Hematology. New York, Paul B. Hoeber, Inc., Vol. II, 1376–1426.

*The blood in disease:*

Coles, J. D. W. A.: 1939. Aegyptianellosis of poultry. Proc. Seventh World's Poultry Cong., p. 261.
Cook, F. W., and Dearstyne, R. S.: 1934. Hematology of the fowl. A. Studies on normal avian blood. B. Studies on the hematology of avian typhoid. N. C. Agr. Exper. Sta., Tech. Bul. 44:51.
Fantham, H. B.: 1912. Blutbeobachtungen bei Waldhühnern. Deut. tierärztl. Wochnschr. 20:247.
Fennell, R. A.: 1947. The relation between age, number, and types of cells in the peripheral circulation of chicken embryos under normal and experimental conditions. Jour. Agr. Res. 74:217.
Gauger, H. C., Greaves, R. E., and Cook, F. W.: 1940. Paratyphoid of pigeons. N. C. Agr. Exper. Sta., Tech. Bul. 62:71.
Grimal, R.: 1938. Variations de la formule leucocytaire et du rapport lymphomonocytaire dans la tuberculose aiguë de la poule. Soc. Biol. (Paris) Compt. Rend. 128:655.
Jackson, C.: 1936. Incidence and pathology of tumors of domesticated animals in South Africa. Onderstepoort Jour. Vet. Sci. and Animal Ind. 6:1–460.
Jacobi, L.: 1939. Beiträge zur Pathologie der Infektion des Huhnes mit Plasmodium gallinaceum (Brumpt). Arch. f. exper. Path. u. Pharmakol. 191:182.
Johns, F. M.: 1934. A study of punctate stippling as found in the lead poisoning of wild ducks. Jour. Lab. and Clin. Med. 19:514.
Johnson, E. P., and Lange, C. J.: 1939. Blood alterations in typhlohepatitis of turkeys, with notes on the disease. Jour. Parasit. 25:157.
Key, J. A.: 1924. Lead Studies, IV. Blood changes in lead poisoning in rabbits with especial reference to stippled cells. Am. Jour. Physiol. 70:86.
Krömker, F.: 1937. Ein Beitrag zum Blutbild gesunder und kranker Hühner. Thesis No. 1840, presented to Friedrich-Wilhelms University, Berlin. R. Pfau, Berlin, p. 41.
Lührs, G.: 1936. Blutuntersuchungen bei kampfstoffvergifteten Tauben, zugleich ein Beitrag zur Morphologie des normalen Taubenblutes. Ztschr. f. Veterinärk. 48:129.
Moore, V. A.: 1895–96. Infectious leukemia in fowls—a bacterial disease frequently mistaken for fowl cholera. Twelfth and Thirteenth Annual Reports. Bur. Animal Ind., U. S. D. A., pp. 185–205.
Olson, C.: 1935. The effect of certain ectoparasites on the cellular elements and hemoglobin of the blood of the domestic chicken. Jour. Am. Vet. Med. Assn. 87:559.
—— and Feldman, W. H.: 1936. The cellular elements and hemoglobin in the blood of chickens with spontaneous tuberculosis. Jour. Am. Vet. Med. Assn. 89:26.
—— and Goetchius, G. R.: 1937. The reaction of chickens to certain members of the colon-paratyphoid group of bacteria. Cornell Vet. 27:354.
—— and Levine, P. P.: 1939. A study of the cellular elements and hemoglobin in the blood of chickens experimentally infected with *Capillaria columbae* (Rud.). Poultry Sci. 18:3.
Pomeroy, B. S., and Fenstermacher, R.: 1937. Hemorrhagic enteritis in turkeys. Poultry Sci. 16:378.
Rostorfer, H. H., and Rigdon, R. H.: 1946. A physiologic study of hematopoiesis in the duck with malaria. Am. Jour. Clin. Path. 16:518.
Seastone, C. V.: 1935. Pathogenic organisms of the genus Listerella. Jour. Exp. Med. 62:203.
Taylor, W. J.: 1916. A report upon an outbreak of fowl typhoid. Jour. Am. Vet. Med. Assn. 49:35.
Thorp, F., and Graham, R.: 1932. Blood-cell counts in acute avian laryngotracheitis. Jour. Am. Vet. Med. Assn. 80:909.
Veenendaal, H.: 1935. Loodintoxicatie en basophiele korreling der roode bloedlichaampjes. Tijdschr. v. Diergeneesk. 62:244.
Ward, A. R.: 1904. Fowl cholera. Calif. Agr. Exper. Sta., Bul. 156.
Wickware, A. B.: 1947. The differential blood picture in chickens before and after administration of embryonated eggs of *Heterakis gallinae* with notes on pathogenicity. Can. Jour. Comp. Med. 11:78.
Wirth, D., and Kubasta, F.: 1939. Studien zur artspezifischen Reaktion der hämatopoetischen Organsysteme (VII, Huhn). Folia Haematol. 62:43.
Yakimoff, W. L., and Rastégaïeff, E. F.: 1929. Sur la question des variations cytologiques du sang des poules. Bul. Soc. de path. exot. 22:766.
——: 1930. Die Spirochätose der Hühner in Nordkaukasus. Zentrabl. f. Bakteriol. I, Originale 117:223.

CHAPTER FIVE

# PRINCIPLES OF DISEASE PREVENTION

By W. R. HINSHAW, *Department of Veterinary Science,*
*University of California, Davis, California*

☆   ☆   ☆

The same principles of disease prevention apply to poultry as to other livestock; and they are even, to a large extent, the same as those applying to human beings. Van Es and Olney (1934) summarize the factors conducive to health and body efficiency: " (1) soundness of body and of constitution and vigor, (2) adequate nutrition, (3) suitable environment, and (4) eradication and control of transmissible diseases." Although immunity to disease cannot be guaranteed when poultry is reared according to these principles, the grower who observes them will increase his chances of raising a profitable flock.

## SOUNDNESS OF BODY AND CONSTITUTION

The most important factor in having a flock of sound, vigorous birds that have good constitutions is the breeding back of the flock. Selection of the healthy, well-matured stock before selling any of the birds for market will aid in building up a disease-resistant flock. The ancestry of the breeding stock should be considered; birds from a parent that had some defect, such as a pendulous crop, crooked toes, or a curved spine, should always be avoided. A chick or poult that has come through the season without any setback will serve much better for propagation than the one that has had several setbacks. Marking the prospective breeders early in the season for later culling to the number needed insures a large group of birds that meet the requirements. Before being finally selected for a breeding flock, each individual should be examined for defects and discarded if abnormal in any way.

When buying hatching eggs or day-old chicks or poults, one should inquire carefully into the source of the stock to be purchased. To meet increasing competition, hatcheries must furnish the kind required by their patrons. The purchaser should demand stock from disease-free breeding flocks that measure up to the principles just discussed.

## ADEQUATE NUTRITION

An adequate diet supplies all the essentials for normal growth. With any one essential food lacking or, even in some instances overabundant, the normal development will be hindered; and a diseased condition, directly or indirectly due to the faulty ration, may result. Whether or not heavy losses from death occur in such cases, slow development may cause as great a monetary loss as if the flock suffered from a heavy mortality. Some of the dietary disorders caused by faulty rations will be discussed under another heading.

## SUITABLE ENVIRONMENT

The term "environment" refers to the surroundings in which the birds must live. Necessarily, this environment varies with the methods of rearing. The practice used in some areas of hatching and rearing with hens on large range areas furnishes an entirely different type of environment from that furnished by the so-called "artificial" method of incubator hatching of eggs and brooder rearing of young birds. Furthermore, the range rearing of poults or chicks is in contrast to the confinement method. In any case, the relation to disease depends on the ability of the environment to aid nature in combating disease. Dryness, drainage, amount of sunshine, nearness to other species of fowl on the same ranch, location in respect to other ranches, type of soil, and shelter facilities are examples of environmental factors that may influence the disease problem.

### ERADICATION AND CONTROL OF TRANSMISSIBLE DISEASES

Transmissible diseases, once established, may cause heavy losses. Examples are blackhead, fowl typhoid, fowl cholera, pullorum disease, paratyphoid, Newcastle disease, laryngotracheitis, fowl pox, and coccidiosis. The two general ways of introducing infectious diseases into a flock are by natural and mechanical carriers.

**Natural carriers.** The most serious carriers of infections are adult fowls or other animals which have apparently recovered from the disease in question but which still retain the infectious organisms in some part of the body where they continue to multiply and to be eliminated. Among the diseases known to be transmitted by apparently normal carriers are blackhead, hexamitiasis, leucocytozoon infection, coccidiosis, fowl typhoid, pullorum disease, salmonellosis, tuberculosis, and fowl cholera. Removing natural carriers from the flock and premises is the most effective way of preventing a recurrence of an outbreak. Different methods of accomplishing this end exist; *but one, common to all diseases, is absolute isolation of the adult breeding flock from the growing flock.*

Disposing of all the birds on the premises and buying day-old poults or

chicks from reliable hatcheries is an excellent method of eliminating natural carriers.

The depopulation method is especially applicable to such diseases as infectious coryza, tuberculosis, fowl cholera, Newcastle disease, and infectious laryngotracheitis. The time between depopulation and buying of disease-free day-old replacement stock will depend on many factors discussed under the diseases in question. Environment plays an important part in the time interval necessary to insure success.

It is extremely important that all replacements for any system of management come as pullorum-disease-free (U. S. Pullorum-Passed or U. S. Pullorum-Clean) eggs, chicks, or poults. No other grades of pullorum certified stock can be guaranteed free of this disease. In fact, such a grade as "Pullorum-Tested" is actually a guarantee that the chicks, poults, or eggs are from a pullorum infected premises. For details about pullorum grades see the chapter on Pullorum Disease.

The purchase of started chicks or poults is an excellent means of introducing many diseases into a flock. Such diseases as Newcastle disease, coccidiosis, and the various respiratory diseases are especially prone to be spread by purchase of chicks that have been reared from a few days to a few weeks by the hatchery.

The purchase of adult birds for breeding flock replacements is another common way of introducing disease into a flock. This practice should not be tolerated. Breeding flock owners should always purchase their replacements as hatching eggs or day-old chicks or poults, and only from known disease-free sources.

An equal chance is taken by the breeding flock owner who exhibits his breeders at shows or fairs and then returns them to his ranch. If he must show his birds, he should select individuals which can be disposed of for market purposes after the exhibition is over. Fowl pox, respiratory diseases, and Newcastle disease are likely to be the price paid for returning birds to the home ranch following their exhibition at shows. Egg laying contests fall in the same category as fairs and shows.

The producer of hatching eggs will have the problem of isolating from his adult flock and other fowl, the chicks or poults kept for replacements.

Unfortunately, carriers of the more common diseases of poultry cannot be detected by simplified tests that are practicable. The agglutination tests for carriers of fowl typhoid and of pullorum disease are exceptions. Only eggs from flocks known to be free of all diseases should be purchased by hatcherymen. Strict supervision over all supply flocks will help to secure eggs of the best quality. Pullorum-free flocks (U. S. Pullorum-Clean or U. S. Pullorum-Passed) only should be maintained.

Chickens may be carriers of many diseases common to both turkeys and

chickens. In some instances—for example, in blackhead—they are fairly resistant, whereas turkeys are highly susceptible. It is equally true that turkeys may be carriers of diseases which may cause severe losses in chickens. Turkeys and chickens can be reared successfully as pen mates or in adjoining yards, provided both species are free of disease; but the chances that chickens may carry blackhead or other diseases or that turkeys may carry chicken diseases are too great to risk (Fig. 5.1).



Fig. 5.1. Chickens and turkeys should not be reared in the same yard. (Hinshaw, Univ. of Calif.)

**Mechanical carriers.** Mechanical carriers include all means by which infectious organisms are accidentally carried from place to place: man, animals, wild birds, insects, dust storms, moving vehicles, and flowing streams.

Man is the worst offender. The attendant who cares for a flock of adult birds that contains coccidiosis carriers is the principal carrier of the disease to young poults or chicks. As experimental work at this station has shown, an attendant may carry coccidia on the soles of his shoes at least one-half mile. Sterilized feed trodden by attendants who have visited yards known to contain coccidiosis carriers has proved to be contaminated and, when given to susceptible birds, has produced fatal cases of coccidiosis. Thus, if adult turkeys or chickens are to be kept on the same ranch with poults or chicks, great care must be taken to prevent spread of the disease from the adults to the young by attendants. This precaution applies also to other diseases.

Visitors, especially other poultry growers, feed salesmen, and service men, are the principal offenders aside from the attendant himself. The poultry

grower should avoid visiting neighboring ranches if disease is known to be present. Visitors should be cautioned about entering the houses and yards. The feed dealer's or the poultry buyer's truck and the borrowed spray tank that has been making the rounds of the ranches may be sources of disease. The used feed sack, the poultry crate that has not been thoroughly cleaned and disinfected after being sent to market, and the hoe or scraper that is used in the pens of carriers and then in the brooder house without being cleaned, are other possible sources of disease.

Since carcasses and offal from birds killed for table use can be classed as possible sources of disease, such material should be burned or buried deep. Contaminated soil, and water polluted by dead birds thrown into streams even at some distance from the ranch, are other sources.

Hospitals and hospital yards may be important in spreading disease to different pens or houses on a ranch. Sick birds from several pens congregated in one hospital pen or house and later taken back to their respective quarters may not only carry back the condition for which they were removed but one or more diseases contracted while in the hospital. For this reason hospital pens are not advocated. Birds removed for treatment should be kept near their respective units.

Wild birds, dogs, cats, rodents, and insects are difficult to incriminate as mechanical carriers, but they are possibilities and should be kept away whenever possible. Certain wild birds susceptible to diseases of poultry are potential natural carriers.

If chickens are reared on the same ranch with turkeys, care should be taken to reduce to a minimum the possibility of infection of one species by the other. An irrigation ditch running from a chicken yard to a turkey yard is a common method of spreading disease. Equally dangerous is the ditch or stream passing through one poultry ranch and flowing through the pens of an adjoining ranch.

## SANITATION

Sanitation may be defined as the means and measures directed toward establishing and maintaining an environment in which it is safe for animals to exist. The factors considered on the preceding pages are important adjuncts to any sanitary program. Especially important is the elimination of carriers. Other factors to consider are houses, yards, water supplies, and food.

**Houses and yards.** The first step in the sanitation of brooder houses is the original construction. Ease of cleaning and disinfection, proper isolation of each unit in case of the multiple-pen type, separate entrances for each unit, sanitary water and feeding systems, and rodent-proof feed storage containers should be considered when building a brooder house. Continuous with the cement floors of the houses, cement yards should be constructed with sides

and with the proper slope to permit cleaning and washing of one pen without danger of getting water and refuse into the adjoining pens. Facilities for cleaning the individual houses and yards can be arranged by having a gate in the front entrance of each yard. A gravel drainage area in front of the yard system or a cement drain to take off the excess water after washing the yards is desirable. Wire sun porches and wire platforms are also an aid in preventing disease (Fig. 5.2) .



FIG. 5.2. Wire sunporches are an aid in preventing disease. This shows an example of a sanitary runway attached to the front of a portable brooder house. (Payne, Kan. Agr. Exper. Sta.)

The number of birds on a given area, either in the brooder house or on the range, may influence the livability percentage to a marked degree. Over-crowding means more work in keeping the surroundings clean and dry; it also increases the problem of feeding and ventilation in brooders. These factors indirectly lower the resistance of birds and facilitate spread of disease.

Yards or ranges should be maintained free of all infections and infestations. Chickens should never be reared in yards with turkeys. For the confinement method of rearing turkeys, rotation of runs is recommended. In large range areas, rotation of runs is impossible; but feeding grounds and feeding areas can be moved at least twice a week as an aid in preventing accumulations of manure and litter, wherein lies the greatest danger of disease. Good drainage that prevents the formation of stagnant pools in yards is necessary. The probability of introducing disease is directly related to the amount of parasitic invasion. If moisture is not present, only the more resistant organisms can remain alive and infective. Good drainage, such as

is found on sandy or gravelly soil, aids in keeping infections at a minimum, because of the dilution factor of rains. Dry, hot regions having an abundance of sunshine aid in reducing the possibility of contamination and therefore in
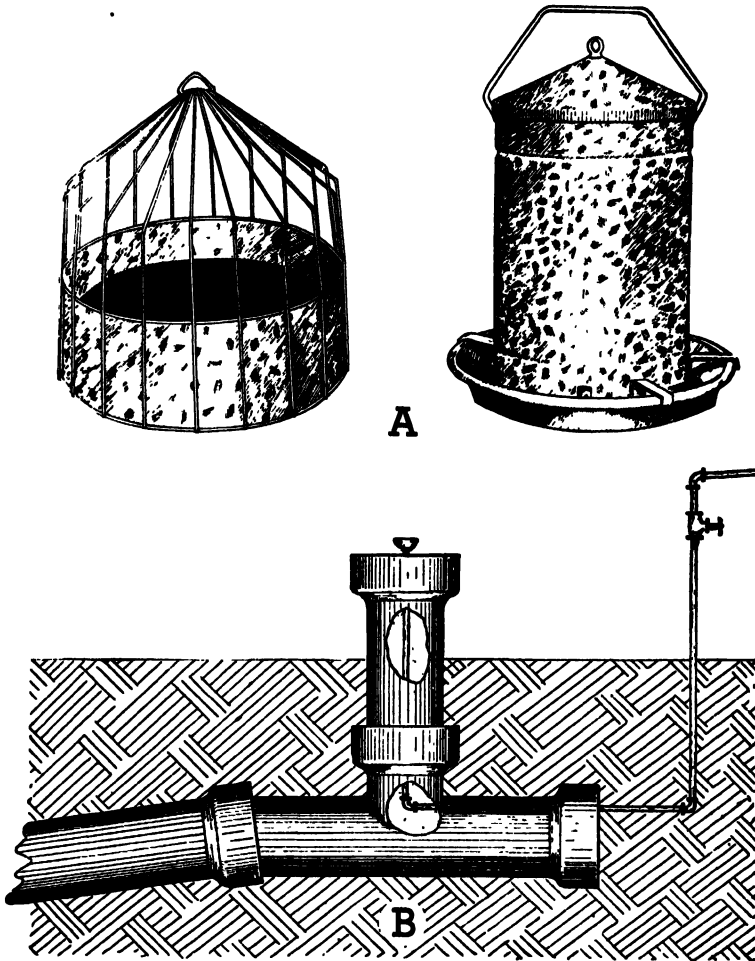


FIG. 5.3. A—two types of commercially made galvanized waterers suitable for poultry. These should be set on wire platforms to insure dry surroundings. B—the "Van Es" type of water fountain. It provides for a continuous flow of water in the bubbler and for passage of overflow into the tile drain. The drinking cup is placed 8 inches above the ground, is kept automatically cleansed, and can be regarded as strictly sanitary. (Van Es, Univ. of Nebr.)

preventing disease. The range method, which provides enough ground so that birds can be moved frequently to clean areas, likewise helps.

**Water supply.** Since the water supply is no better than the poorest water available, all sources other than those known to be clean and safe from contamination should be removed. The best type of water fountain is of no

value in preventing disease if it is allowed to overflow and to form a stagnant pool. The immediate area around the permanent fountain or drinking place should be filled in for several inches with gravel, or the container should be set on a screened platform to insure a dry area, which will help to prevent the spread of disease. In houses or in yards, wherever possible, an automatic watering system with proper drainage for disposal of the surplus is recommended. Figure 5.3A and B illustrates types of watering devices that are recommended. Many other types of ready-made sanitary water equipment are on the market.

Streams and irrigation ditches as a source of water are safe provided they come from uncontaminated sources, are not stagnant, and are flowing at a fair rate of speed. *Pools of stagnant water from overflowing or leaking canals or water from ditches that are not flowing cannot be considered reliable.* Since poisoning from salt water and alkali water has been reported, such waters should always be avoided.

Pure, fresh, clean water is the most palatable. If well protected from contamination by body wastes, soil, and feed, it far surpasses the same water which has been medicated. Birds do not like most of the common antiseptics recommended for drinking water; often they avoid water because of this dislike. Frequent changing of water or the use of a sanitary drip or cup system is usually preferable to the use of antiseptics.



Fig. 5.4. A—a practical type of metal feeder designed for hanging on the outside of the wire fence enclosure of a wire-floored sun porch. It can be filled without going into the pen. B—a type of metal feed hopper with wire guards that aid in keeping the feed clean. Note that this type of feeder can be raised or lowered to accommodate different sizes of birds. (Hinshaw, Univ. of Calif.)

**Feeds and feeding methods.** Feed as a mechanical means of carrying infection has already been mentioned. In addition, feed may directly transmit fungus diseases, botulism, and possibly paratyphoid infections. For these reasons, one should purchase the best feed and protect it from dampness and from all sources of infection. Moldy feed should never be given.

Several outbreaks of mycosis originating from contaminated milk containers have been observed by the author. Failure to wash and scald daily

the cans used for transporting milk from the dairy to the range has been the most common cause. Improper care of semisolid milk has been another source of mycosis.

Safeguarding the feed against fecal matter and other refuse by using properly constructed feed hoppers is a necessary procedure in the sanitary program; and cleaning the hoppers scrupulously at frequent intervals is important. *Sweepings from the floor of the feed house should never be given to young growing stock,* because of the danger of refuse carried from the adult flock on the attendant's clothing and shoes. If feed is mixed on the ranch, mechanical mixing is much superior to hand-mixing and reduces the chance for accidental contaminations. Feed hoppers that aid in preventing disease are illustrated in Figure 5.4A and B. Many suitable types of metal hoppers are on the market.

Excess feed scattered around yards attracts rats, mice, and birds, which are all potential carriers of disease (Fig. 5.5). Proper storage of feed in rat- and mouse-proof bins will thus aid in disease control. Sacked feed that must be stored can be kept reasonably free from rats and mice by carefully rat-proofing storage houses (Silver, Crouch, and Betts, 1942) (Figures 5.6, 5.7, 5.8). Storer and Mann (1946) present a comprehensive bibliography of rodent control. Metal garbage cans with covers make suitable bins for temporary storage of feed in brooder houses and in yards. These should be placed outside of the yards whenever possible (Fig. 5.9).

The liberal use of wire platforms for waterers and feeders will do much to aid the program of sanitation. When used, the size should be such as to allow ample room for the equipment and wide enough so that the entire bird can get on it. The height is also important to insure plenty of room for droppings to collect. Figure 5.11 shows a type of platform which has proven very satisfactory for yards and houses.

## DISINFECTION AND DISINFESTATION

*Disinfection.* Disinfection is important in reducing the amount of infection in flocks where carriers exist and, after an outbreak of disease, in destroying the enormous numbers of parasites eliminated. Disinfection at frequent intervals during an outbreak helps to reduce the amount of infection and thus to prevent further spread. It may be accomplished by mechanical, physical, or chemical means. Success depends on the nature of the environment, the character of the infectious agent to be destroyed, and the method to be used. The procedure must fit the special conditions on the ranch. In most cases a combination of the three means gives the best results.

Cleaning, before the final application of chemicals, is essential in any disinfection program; cleaning alone will not result in disinfection, but, if carried out properly, it will render disinfection by chemicals more efficient.
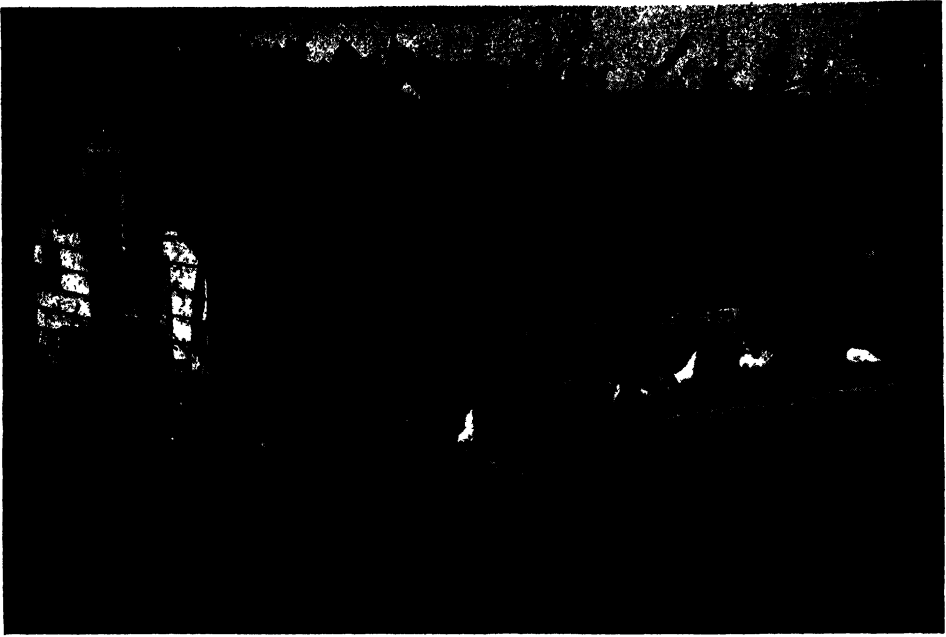
Fig. 5.5. The type of surroundings that invite rat and mice invasions, and in turn increase disease transmission possibilities. (Hinshaw, Univ. of Calif.)
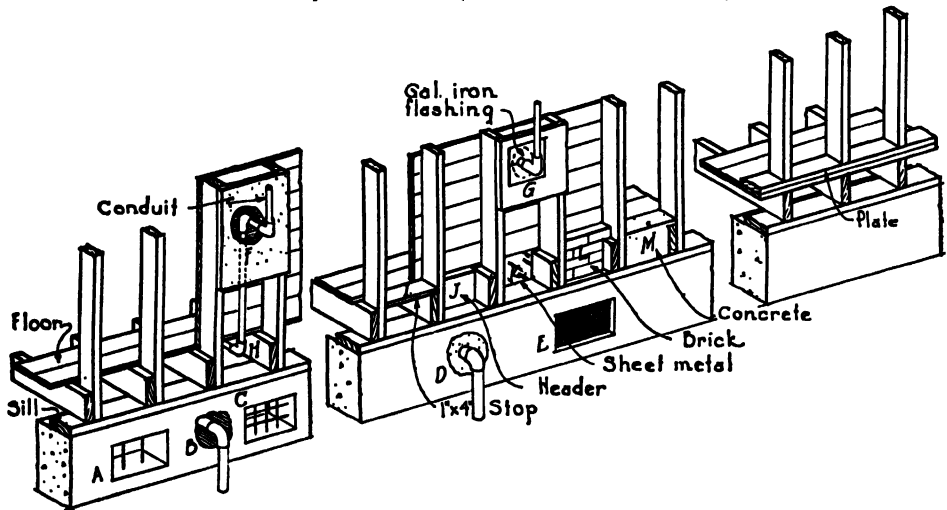


Fig. 5.6. Structural details of buildings in relation to exclusion of rats and mice. Undesirable features: *A,* ventilator to basement with widely spaced bars; *B,* space about entrance of pipe; *C,* ventilator with wide-mesh grille; *F,* hole in wall around entrance of conduit; *H,* free passage for rats from below the floor into the walls.

Corrective measures: *D,* filling space around entrance of pipe with concrete; *E,* covering ventilator with hardware cloth of ⅓- or ¼-inch mesh; *G,* covering entrance of conduit with close-fitting sheet-metal flashing; *I,* stop of wood at floor level; *J,* header block between joists, completely closing the space between sill and floor; *K,* sheet-metal "header"; filling space between studs with brick *L,* or concrete *M.*

At the right is shown the usual "western" type of framing which, when walls are in place, prevents rodents in a basement from gaining access to spaces between studs and walls. (Storer, Univ. Calif., Agr. Ext. Cir. 79.)

Water or weak soapsuds used to flush the walls of a brooder house may serve, however, as a medium for growth of organisms and, unless collected and not allowed to run over the ground, may spread infection. The following steps are suggested for cleaning and disinfecting brooder houses and cement runs before chicks or poults are put in them:

1. Settle the dust by spraying lightly with the disinfectant to be used. This procedure avoids undue scattering of bacteria by the dust.

2. Haul all litter and droppings to a well-isolated portion of the ranch where there will be no danger of contact with poultry. Never spread litter or droppings on the land being used for ranging turkeys or chickens. If infection is known to exist, burn the litter.

3. Remove all movable equipment to the cement run or to a cleaning floor or platform if one is available.

4. Scrub the walls, floors, and equipment with hot lye solution made by adding 1 pound of lye to 20 gallons of hot water. An old broom can be used to apply the lye solution; care should be taken to prevent the fluid from getting on the hands and face. About 1 hour after its application, the lye should be rinsed off with hot water.

5. Spray the walls, floors, and all equipment with a good disinfectant of the concentration recommended by the manufacturer. Use a compressed air sprayer for applying the disinfectant, and cover every part of the building or equipment.

6. Allow time for drying before using the house and equipment again.



FIG. 5.7. Sectional view through a door showing use of sheet metal around edges of door (sides as well as top and bottom), and on the stop and sill to prevent entrance of rats and mice. Maximum clearance, $\frac{3}{8}$ inch. A sill abutting *against* the door is preferable to a sill *beneath* the door. (Storer, Univ. Calif. Agr. Ext. Cir. 79.)

Many poultrymen, turkey growers, and hatcherymen use portable high pressure steam cleaning units for cleaning houses and equipment. Used properly, these units are of great value in the cleaning and disinfection program. Many types are available. One such unit is illustrated in Figure 5.10.

Certain types of hovers and brooder-heating equipment are not easily washed and disinfected by this method because of the danger of injuring

them. The formaldehyde gas method recommended for disinfecting cabinet types of incubators may be used for such equipment if a gas-proof room is available.

After the poults or chicks are put in the brooder house, frequent dry cleaning without disinfection is sufficient. This procedure is recommended because moisture promotes the development of coccidia.

**Disinfestation.** Mechanical or physical means of hindering the development of parasites or destroying them are probably as important as chemical means. Cleaning the yards of all refuse, removing litter and droppings frequently, and constructing the houses so as to prevent the harboring of ticks, lice, and mites are examples of mechanical methods. All methods of fly control—trapping, cleanliness, and removal of breeding places—indirectly aid in reducing tapeworm infestation. Oil sprays for mite and tick control, nicotine sulfate or sodium fluoride for lice, and the use of DDT for fly and mosquito control are chemical means of destroying parasites.
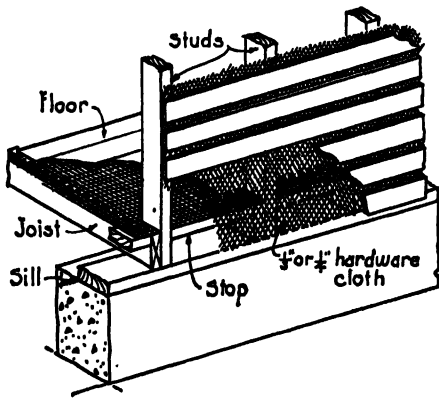


FIG. 5.8. Construction for corn crib to exclude rats. The ends of studs and joists are nailed together to resist lateral pressure when the crib is filled. Spaces between studs are closed by wooden stops, at floor level. Hardware cloth is placed between joists and flooring and between studs and outside slats. The whole crib is elevated on concrete, field stone, or wooden sills so that the floor is at least 12 inches above ground. (Storer, Univ. Calif. Agr. Ext. Cir. 79.)

The methods recommended for cleaning and disinfection are also applicable in the disinfestation program. Yards are best treated by frequent cleaning and by rotation. The former dilutes the amount of infection or infestation and allows the sun better opportunity to exert its influence on the remaining parasites. Rotation of runs at regular intervals allows the sun and the other natural elements to free a given area of parasites and bacterial forms. No satisfactory cheap disinfestant for the soil has been found.

Plowing of the yards is not recommended unless necessary for weed control or unless a crop-rotation system is combined with the poultry-rearing program. Plowed yards soon become dusty, tend to become jutted with holes that collect water during rains, and are harder to clean than yards that are left unplowed.

### DISINFECTANTS

The number of chemicals sold as disinfectants is great. Some are worthless; others are excellent disinfectants but have undesirable characteristics.

FIG. 5.9. Metal cans with tight covers make suitable rat-proof feed containers. They should be placed outside the yards whenever possible. (Hinshaw, Univ. of Calif.)



FIG. 5.10. A portable steam cleaner in operation at the Poehlmann Hatchery, Petaluma, California. (S. E. Hall Co., Berkeley, Calif.)

Among the properties of an ideal disinfectant are (1) low cost per unit of disinfecting value, (2) ready solubility in hard water, (3) relative safety to man and animals, (4) efficient deodorization, (5) easy availability, (6) non-destructibility to utensils and fabrics, (7) stability when exposed to air, (8) absence or minimum of objectionable and lingering odor, (9) effectiveness for a large variety of infectious agents. Obviously, no one chemical will have all these properties; but the list will serve as a guide.

Many disinfectants of similar composition are sold under different trade names. Before buying a product under an unfamiliar trade name, one should
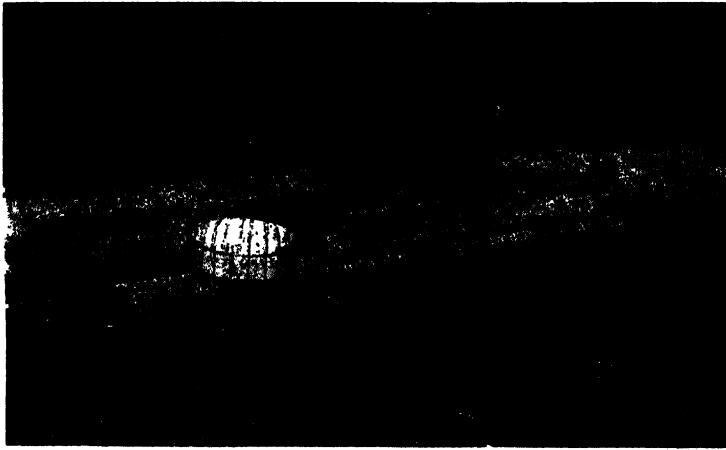


FIG. 5.11. Type of wire platform used at the Los Angeles County (California) Poultry Demonstration Plant, for feeders and waterers. Note the length, width, height, and the size of the wire mesh (1″ x 1½″). (Hinshaw, Univ. of Calif.)

compare types and values with a well-known product. The directions for dilution given by the manufacturer should be followed in making up a disinfectant for use. These directions are usually based on the concentration of the product; and by comparing the dilution factor of two disinfectants that have other properties equal, one can determine the relative cost of the two. For instance, if one disinfectant can be used at the rate of 1 part to 40 parts of water, while another has to be used at the rate of 1 part to 20 parts of water, the first, other things being equal, is worth twice as much as the second.

**Phenol, or carbolic acid.** Phenol is a chemical substance obtained from coal tar. In its pure form it occurs as colorless needles having a characteristic odor familiar to everyone. It is usually sold in water solutions and is too expensive for general use. A 2 per cent solution is a useful antiseptic for wounds; but stronger solutions, as a rule, are caustic. This is the chemical used as a basis for determining the phenol coefficients of disinfectants.

**Crude carbolic acid.** Crude carbolic acid is a mixture of phenol, cresol,

and certain impurities. Its usefulness varies directly with the percentage of cresol, which has a higher disinfecting value than phenol. As its composition is uncertain, it cannot be classed as a desirable disinfectant for general farm use. It is sometimes used in oil mixtures for controlling mites and lice in poultry houses; but since the oil is as effective without the crude carbolic acid, there is no reason for combining the two.

**Cresol.** Cresol is a thick yellow or brown liquid, miscible with water but only slightly soluble. It forms the basis for a large number of the best commercial brands of disinfectants, made by combining cresol with a soap base.

Compound solution of cresol (liquor cresolis compositus U.S.P.), the most refined of the saponated cresol solutions, is composed of cresol 500 grams, linseed oil 350 grams, potassium hydrate 80 grams, and water to make 1,000 grams. Saponated cresol solutions are more effective and less toxic than phenol, can be used in low percentage solutions, are reasonably priced, and are fairly stable in the presence of organic matter; but they have the disadvantage of being soapy and of having the odor characteristic of the cresols. They can be recommended for general use on the farm.

Disinfectants used should conform with the specifications of the United States Department of Agriculture, Bureau of Animal Industry (Mohler, 1940). Saponated cresol solutions applied under supervision of the Bureau must contain not less than 50 nor more than 53 per cent total phenol and not less than 21 per cent by weight of soap; they must form clear solutions when mixed with water, and must be used in the proportions of 4 fluid ounces per gallon of water.

**Coal-tar disinfectants (sheep-dips).** Coal-tar disinfectants are cresol products that form milky emulsions when mixed with water. They vary greatly in their solubility and disinfecting value and should be diluted according to the directions given by the manufacturer.

**Chlorine gas.** Chlorine gas is the basis of the disinfectants known as hypochlorites. The numerous brands of these products offered for sale vary in their disinfecting value according to their chlorine stability and their ability to liberate chlorine gas. They should contain at least 2.6 per cent by weight of available chlorine, the active disinfecting element of such products. Such solutions, if used according to directions, are highly efficient. Their chief disadvantage is the instability of the chlorine when exposed to air or organic matter. They are also quite expensive. Their principal use is for disinfecting limited areas such as incubators, small brooders, and water and feed containers. *All surfaces to be disinfected with hypochlorite solutions must first be thoroughly cleaned in order to insure the greatest efficiency. Stock supplies should be kept in dark cool places, and the containers should be tightly sealed when not in use.*

**Chlorinated lime.** Chlorinated lime, known as bleaching powder, is pre-

pared by saturating slaked lime with chlorine gas. It should contain from 30 to 35 per cent of available chlorine. The Bureau of Animal Industry recognizes chlorinated lime containing at least 30 per cent available chlorine for official disinfection when used in proportions of 1 pound to 3 gallons of water. Products containing less available chlorine should be used in more concentrated solutions. The final dilution should contain approximately 1.2 per cent of available chlorine by weight. Fresh solutions must be prepared daily. All products containing chlorine must be handled with care; because free chlorine is destructive to fabrics, leather, and metal.

The use of chlorine on the poultry ranch is limited to disinfection of drains, water containers, and feed containers. Its instability makes it of doubtful value for general disinfection.

**Quicklime (unslaked lime, calcium oxide).** The action of quicklime depends on the liberation of heat and oxygen when the chemical comes in contact with water. On the poultry ranch its use is limited to small yard areas that are damp and cannot be exposed to the sun, to the disinfection of drains and fecal matter, and to whitewashes. Adding chlorinated lime to quicklime at the rate of 1 pound to 40 gallons of wash increases its disinfecting value in whitewashes. As quicklime has a caustic action, birds should be kept away from it until it has become thoroughly dry.

Hydrated lime, according to Yushok and Bear (1944), when used as a preservative and deodorizing agent for poultry manure also has value as a partial disinfectant. Mixed with fresh manure at the rate of 200 pounds per ton of manure, it was found to have a bactericidal effect on *Salmonella pullorum, Salmonella typhimurium, Salmonella gallinarum,* and *Pasteurella avicida* in a 15-minute period. Similarly it prevented the sporulation of coccidial oocysts, and the segmentation and embryonation of *Ascaridia lineata* eggs. Another advantage of this use pointed out by them is that the treated manure is unattractive to flies and rodents. Fly maggots are not produced in the treated dropping pits, and both mice and rats avoid them.

**Lye.** Lye is an excellent cleansing agent, valuable in any disinfecting program. A 2 per cent solution of sodium hydroxide (soda lye) is a good disinfectant for many of the pathogenic microorganisms. Because of insufficient data on its disinfecting value against some of the common poultry disease-producing organisms, however, it should be used primarily as a preliminary cleansing agent. Being a severe caustic, it should be handled with care.

**Formaldehyde.** Formaldehyde is a gas, sold commercially in a 40 per cent solution with water under the name of formalin. For spraying it is used in a 10 per cent solution of formalin (that is, a 4 per cent solution of formaldehyde). Though a powerful disinfectant, it has many disadvantages, especially its volatility, penetrating odor, caustic action, and tendency to harden the skin—properties which make it disagreeable to apply. Its chief advantages

are as follows: (1) it can be used as a gas or vapor for fumigation of incubators or small rooms; (2) it is relatively nontoxic to animals and fowls; (3) it is an efficient disinfectant in the presence of organic matter; and (4) it does not injure utensils and spraying equipment with which it comes in contact.

Its use on the turkey or chicken ranch is limited to disinfection of brooder equipment, incubators, water and feed containers, and occasionally—during outbreaks—fumigation of clothing and small utensils that are difficult to disinfect by other means. Fumigation of brooder houses with formaldehyde is, as a rule, impractical because of the difficulty in making them airtight.

Fumigation of incubators and incubator rooms is a practical procedure, in common use by hatcherymen. Most manufacturers have recommendations for their type of machine; and, when possible, their directions should be followed. When fumigating a room or an incubator, one must have the space airtight and the room temperature and humidity as high as possible. For most efficient disinfection of incubators, Bushnell and Payne (1931) recommend a wet-bulb thermometer reading of 85° 'to 95° F. Disinfection is uncertain in rooms having a temperature of less than 65° and a relative humidity of less than 60 per cent.

The two common methods of fumigating cabinet-type incubators (forced- or circulating-air types) are given below:

1. Formaldehyde gas is generated by mixing formalin (40 per cent formaldehyde) and potassium permanganate. For this purpose 35 cc. (1.2 ounces) of commercial formalin and 17.5 grams (0.6 ounce) of potassium permanganate for each 100 cubic feet of incubator space are mixed together in an earthenware or enamelware vessel having a volume of four to five times the amount of material used. The vessel should be placed above the floor in the middle compartment of the incubator (Fig. 5.12). The doors should be kept closed for at least 10 minutes to allow the gas to penetrate to all parts of the machine. Equipment for generating and introducing the gas through the intake parts of certain types of machine is obtainable from the manufacturers.

Insko et al. (1941) recommend using from two to three times the normal amount of potassium permanganate and formalin between hatches when omphalitis is being transmitted in an incubator.

Compartment-type (still-air type) machines do not lend themselves to fumigation methods as well as do the cabinet types, but can be so disinfected by opening them and fumigating the room in which they are located.

2. Formaldehyde gas liberated from formalin-soaked cheesecloth is recommended by the Illinois Agricultural Experiment Station (Graham and Michael, 1933). Before disinfection, the incubator must be thoroughly dry-cleaned. Approximately 20 cc. of formalin is used for each 100 cubic feet of

incubator space. A saturated cloth large enough to carry the formalin without dripping is suspended under or near the circulating fans, and the formalin allowed to evaporate (Fig. 5.13A and B). This method is said to be as efficient as the first and is less expensive.

Formaldehyde can be used successfully when eggs are in the incubator.



Fig. 5.12. A method of fumigating an incubator with potassium permanganate and formalin. (Graham and Michael, Univ. of Ill., 1933.)

Insko *et al.* (1941) warn against fumigation during the first three days of incubation because the embryos are then most susceptible to formaldehyde. It is of especial value for disinfecting between hatches in large incubators when chicks or poults are hatching at short intervals of time. Fumigation with formaldehyde destroys or attenuates the pathogenic organisms in the incubator, but not within the egg nor within the body of the hatching chick or poult. Its principal use, therefore, is in disinfecting incubators between hatches and, in some instances, during the early stages of a hatch. Before fumigation of incubators during the hatching period, advice should be sought from one familiar with the procedure to determine the possible need and methods. In general, fumigation of hatching chicks or poults is not recommended.

**Copper sulfate (bluestone).** Although copper sulfate and other salts of copper have a marked toxic effect upon some of the lower forms of life, they are not considered good general disinfectants. Copper sulfate is effective against algae and certain fungi and may prove of some value in outbreaks of fungus diseases. Copper sulfate of a greater concentration than 1 part in 500
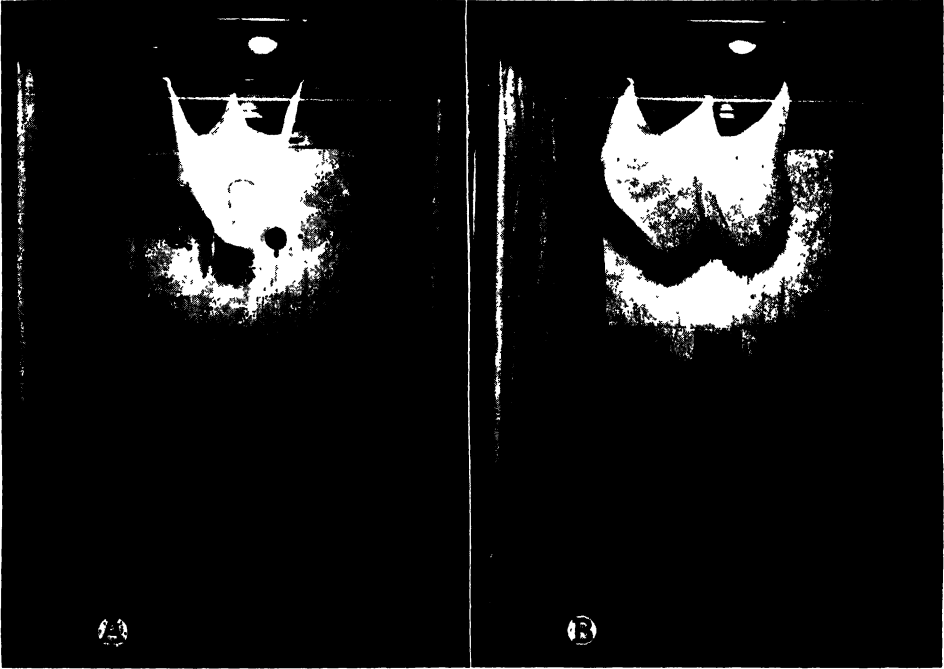


Fig. 5.13. A method of fumigating an incubator with formalized cheesecloth. (A) Formalizing the cheesecloth. The formalin is measured into a small container, such as a half-pint milk bottle. Then the cloth is thrust into the formalin and the container inverted. The formalin is absorbed almost instantly. The formalized cloth is then removed from bottle and hooked under the screened fan in a hammock-like manner. (B) Cheesecloth suspended under fans after it has been formalized. (Graham and Michael, Univ. of Ill., 1933.)

of water may be toxic when given as the only source of drinking water. Turkeys do not like copper sulfate solutions of any concentration and will seek other water supplies if they are available. A 0.5 per cent solution may be of value for disinfecting feed hoppers, water fountains, and areas around these in fungus-disease outbreaks.

A convenient method of making up approximately a 1:2,000 dilution of copper sulfate solution is given below.

Stock solution: Dissolve 1 pound of copper sulfate (bluestone) in 1 gallon of soft water (rain water or distilled water). If soft water is not available add 1 teaspoon of concentrated hydrochloric acid or 1 cup of vinegar to the water

before adding the copper sulfate. It may be necessary to heat the mixture to dissolve the copper sulfate. Store in a glass bottle.

To make a 1:2,000 dilution: Add 1 tablespoon of the stock solution to each gallon of water. It is necessary to acidify hard water by adding just enough vinegar or hydrochloric acid to prevent precipitation of the copper. The amount of acid will vary with the hardness of the water. Not over 1 teaspoon of hydrochloric acid should be added to each gallon of water.

Copper sulfate should not be used either for general disinfecting purposes or for use in drinking water except when recommended for controlling a specific disease which has been definitely diagnosed.

**Potassium permanganate.** Potassium permanganate depends on its rapid oxidizing property for its disinfecting value. Although it has little usefulness as a general disinfectant, certain properties make it convenient as an antiseptic for drinking water. It is inexpensive, and when it has lost its disinfecting power, it turns brown. As it corrodes metals, it must be used in earthenware or wooden vessels. One level teaspoon of potassium permanganate for each gallon of water will aid in reducing the chance for the spread of disease, but it has no medicinal value. Potassium permanganate solutions lose their antiseptic power so quickly in the presence of organic matter that they are useless for mouth or crop treatment.

**Sodium orthophenylphenate.** This substance has only recently been recommended as a general disinfectant. It has no objectionable odor, is relatively nontoxic, is highly efficient for most pathogenic microorganisms, and is readily soluble in water. It may be purchased in the form of a grayish, brownish, or white powder or flakes, which must be kept in a closed container to prevent deterioration. It is now sold under several trade names, which are included under the Bureau of Animal Industry list of permitted disinfectants. It gives best results when applied hot. According to trials made by the United States Department of Agriculture, it is effective for mite and lice control.

**Iodine.** This disinfectant, though effective, is too expensive for general use. Tincture of iodine is a valuable antiseptic for skin wounds but should not be used internally.

**Mercuric chloride (bichloride of mercury, corrosive sublimate).** Although a powerful disinfectant, mercuric chloride is limited in usefulness by its cost, toxicity, and marked corrosive action on metals. It is commonly used in a 1:1,000 dilution with water. Because its value is markedly lowered by the presence of organic matter and because it has certain other undesirable properties, it cannot be recommended for disinfection of litter or houses. Many other disinfectants, almost as efficient and less poisonous, are preferable for use on the poultry ranch.

**Quaternary ammonium compounds.** Within the past few years much

publicity has been given to the disinfecting value of quaternary ammonium salts. There are a number of these products now on the market, and they are generally considered to be good disinfectants if used according to directions. They are water clear, odorless, are nonirritating to the skin, and have a marked detergent (cleansing) action. These compounds are recommended especially for disinfection of eggs, and for general use around the hatchery. It is important to remember that quaternary ammonium compounds cannot be used in soapy solutions.

**Glycol compounds.** The germicidal action of glycols on air-suspended bacteria and viruses was first demonstrated by Robertson *et al.* (1941). Attempts to adapt them to use for incubator air disinfection have been made by several investigators, including DeOme (1944, 1946) and Gwatkin (1947). Direct application to hatchery use was studied by Gwatkin, who concluded that propylene or triethylene glycol have little value as incubator disinfectants.

**Sunlight.** The sun's direct rays are the best disinfectant known. Since, however, the material to be treated must be in thin layers and exposed to the direct rays, this method of disinfection is limited to yards and to utensils that can be thoroughly cleaned before being exposed. The construction of most poultry houses prevents efficient disinfection by the sun. A cement platform fully exposed to the sun makes a convenient place for treating movable equipment. If properly constructed with a drain, such a platform can be utilized as a washing and disinfection rack.

There are many types of germicidal lamps now being advertised for use on the poultry farm and in the hatchery, but there is not enough scientific evidence available to warrant a recommendation for their general use.

**Hot water.** Hot water adds to the efficiency of any disinfectant and, if applied in the form of boiling water or live steam, is effective without the addition of any chemical. Live steam must be applied directly to the part to be disinfected (see Fig. 5.10).

**Dry heat.** Dry heat in the form of a flame is effective provided the flame comes into contact with the bacteria to be killed. According to experiments by Stafseth and Camargo (1935), the fire guns commonly used on poultry farms are not highly efficient as a means of disinfection. They may be of some value in drying up the floors and walls after the use of watery solutions of disinfectants and also in drying up damp areas around water and feed containers after a dry cleaning of brooder-house floors. *All methods involving direct flame are, however, dangerous fire hazards.*

## DISINFESTANTS

Disinfestants, sometimes called parasiticides, destroy animal parasites such as lice, mites, ticks, and fleas. Their use is recommended only as an adjunct

to a properly conducted sanitary control program. Many disinfectants are also destructive to lice, mites, and other similar parasites, provided they come in contact with the parasite. Many, however, are useless as disinfestants.

**Crude oil, distillates, and similar cheap oils.** Petroleum oils are excellent and cheap agents for the destruction of lice, mites, and ticks. There is no advantage in adding disinfectants to oils for lice and mite control; the oil itself is effective, and oil lowers the value of a disinfectant. Since the oil must come into direct contact with the parasite, refuse must be removed and all hiding places made accessible before the application.

Carbolineum and similar products which contain anthracene oil have good penetrating properties, and are very effective for controlling mites and lice within the poultry house. Creosote oils of the penetrating types used for termite control are helpful in lice, tick, and mite control if not too expensive.

Bullis and Van Roekel (1944) have reported that exposure of chicks to the fumes of coal-tar creosote oil, anthracene oil, and certain mite paints too soon after use in a brooder house may cause anasarca (ascites, watery belly).

**Nicotine sulfate.** A 40 per cent solution of nicotine sulfate, such as is sold under the trade name of Black Leaf 40, is in general use for control of lice on chickens. Its action depends on a volatile substance that penetrates the feathers of the birds when it is painted on the perches just before they go to roost. The method is not well adapted to control of lice on turkeys under rearing conditions where the perches are usually placed out of doors. Turkeys may be treated by applying the nicotine with a small brush to the feathers over the parts most often affected with lice. Some turkey growers use a small oil can as a dispenser for the drug, and apply a drop on the feathers.

**Sodium fluoride.** This, either as a dust or as a dip, is effective for ridding birds of lice. The dusting method is probably the most desirable. It consists of rubbing a pinch or two of the powder into the parts most often infested with lice (on the tail, under the wings, on the neck and head, and on the breast). Such treatments should be repeated in about two weeks if the birds are badly infested.

**DDT (dichloro-diphenyl-trichloroethene).** This insecticide is not a panacea for the control of external parasites, but it has a definite place in the disease prevention program. Its effectiveness has been established for killing flies, mosquitoes, and the poultry tick. It is of no more value for lice or mite control than are some of the other well-known remedies, so until further information is available it is suggested that its use be confined to the control of flies, ticks, and mosquitoes.

There are five general types of product containing DDT available com-

mercially. They are as follows: (1) solutions in kerosene or similar solvents; (2) dusting powders; (3) wettable powders to be mixed with water; (4) emulsion concentrates to be mixed with water; and (5) aerosol bombs for space spraying. For use on the poultry farm, wettable powders are probably the most usable for general application. Wettable powders contain a wetting agent and are sold with DDT concentrations varying from 20 to 50 per cent. Under most farm conditions good results will be obtained with a 2.5 per cent suspension of DDT in water. The interval between applications will depend on the amount of residue left on the sprayed surface, and on environmental conditions. The criterion for the determination of this interval should be the existing population of insects on the sprayed premises.

In the amounts needed for control, DDT is safe to use in brooder houses and laying houses. Water emulsions are less apt to be toxic than are oil sprays. The fact that the chemical is toxic when used in sufficient amounts makes it necessary to use it with caution, especially in houses that are occupied.

## REMEDIES

The most successful poultry growers are those who feed adequate diets consisting of simple ingredients, give the birds all the fresh, pure water they will drink, and spend little or no money for proprietary remedies. The number of specific and useful remedies is limited.

Recent research work indicates that certain of the sulfonamides may have value for the treatment of coccidiosis, fowl cholera, and the salmonelloses. Details concerning their uses and value are given under the specific diseases. It should be emphasized that none of the reports published to date show that these drugs eliminate the carriers from the flocks, nor do they take the place of the sanitation program. Penicillin has shown promise as a treatment for erysipelas, and spirochaetosis. Much needs to be done with all of these remedies before they can be generally recommended, and each case must be carefully considered before treatment is recommended.

The periodic use of laxatives such as Epsom salt is of questionable value.

A flock should never be treated for any species of parasites until a large number of the birds have been examined. A few parasites in one or two birds do not justify treatment. In no case should tapeworm remedies be given to the entire flock before testing out the drug on a few birds. This is necessary because most tapeworm remedies contain kamala, which varies in its toxicity, especially for turkeys.

## HANDLING AN OUTBREAK OF DISEASE

A daily inspection of the flock is essential. At the first signs of the birds becoming droopy, losing their appetite, or in any way appearing abnormal,

one should start looking for the possible cause. Every disease outbreak should be considered infectious until the contrary is proved. From the first appearance of abnormalities in a flock, the following suggestions should be observed:

1. Isolate all abnormal birds. The best method is to remove the healthy-looking individuals and put them in cleaned and disinfected quarters or on ground that has not been used for poultry for several weeks. Be sure that all the feed and water containers are thoroughly cleaned and disinfected before being transferred to the new quarters.

2. If the cause cannot be readily determined, a few of the typically sick specimens should be submitted to a diagnostic laboratory.

3. Advise the owner to kill hopelessly sick birds by breaking their necks to avoid shedding of blood and thus prevent the spread of infections that are present in the blood stream. A convenient tool for the purpose is a Burdizzo forceps like that used for castrating calves (Fig. 5.14). Another means of killing birds for autopsy is by electrocution.



Fig. 5.14. Use of Burdizzo forceps in killing a turkey. When brought into a closed position carefully the jaws separate the vertebrae and sever the spinal cord and jugular vein without breaking the skin. (Hinshaw, Univ. of Calif.)

4. Burn or bury dead birds. If buried they should be placed deep enough to insure their not being dug up by dogs or other animals.

A disposal pit such as is illustrated in Fig. 5.15 is superior to incineration as conducted on most poultry farms and by hatcheries. Such pits are easily and cheaply constructed, and are very efficient. The roof of such pits and especially the "manhole" covering must be air tight to prevent escape of odors and avoid the attraction of flies. Periodic spraying of the roof of the pit with DDT emulsion is suggested. Quicklime may be used to hasten decomposition, but is not necessary. Open disposal pits are not recommended.

5. Thoroughly clean and disinfect all houses and equipment. If the affected birds are in yards, these should be cleaned of all refuse to allow the sun to aid in disinfecting all parts.

6. Keep fresh water before the birds at all times. The water containers

should be thoroughly washed and disinfected at least once daily. Antiseptics in the drinking water are of questionable value and may cause them to seek other sources of water supply much less desirable than pure untreated water. Stagnant pools or irrigation ditches should be fenced off so the birds cannot use water from them.

7. Thoroughly clean and disinfect all feed hoppers daily.

8. Thoroughly inspect the food to determine the possible presence of decayed fish or meat scraps, spoiled milk, moldy grain, poisonous weeds, or other sources of possible trouble.

9. Avoid sudden changes of feed. If the feed is the cause of the trouble, a new diet is warranted; but any changes should be made by gradually increasing the new formula. As a rule, reduction of the protein level of the ration is desirable during an attack of enteritis. The addition of bran to increase the bulk of a feed and the use of a so-called meal method of feeding will often aid in stopping a mild case of enteritis which might cause severe losses if untreated. In these



FIG. 5.15. Poultry disposal pit. Such a pit can be made any size that is convenient, and is valuable for disposal of hatchery wastes as well as carcasses. (Hinshaw, Univ. of Calif.)

cases the mineral-oil treatment given below is also of value.

10. If diarrhea is present, administer a mild laxative, such as a medium or light grade of mineral oil given at the rate of 2 quarts per 100 pounds of mash or bran for a period of 3 days. If Epsom salt is used, the amount should not exceed 1 pound to each 1,000 pounds of turkeys or 500 pounds of chickens. Milk flushes, though satisfactory for certain disease conditions, may cause severe losses in others. It is not desirable, therefore, to give milk in quantities for medicinal purposes until a definite diagnosis has been made. Molasses added to a bran mash or to the regular mash at the rate of 5 per cent of the ration acts as a light laxative and is relished by the birds. This amount of molasses should be considered a remedy and not given for over 2 or 3 days at a time (see paragraph 11).

11. The convalescent stage of any disease is the most important one. Poults or chicks die from lack of feed and water in a very short time.

Getting them to eat after they have been ill, even for a short time, is often a very difficult task. It must be accomplished, however, if the mortality is to be reduced to a minimum. Feeding small quantities of mash at frequent intervals often aids in restoring feeding habits. Milk products if used judiciously are good convalescing foods. The use of small amounts (not to exceed 5 per cent) of molasses in bran or bran plus the regular mash, sometimes serves as an appetizer if other methods fail. Plenty of fresh water and succulent greens should be given.

## REFERENCES

Bullis, K. L., and Van Roekel, H.: 1944. Uncommon pathological conditions in chickens and turkeys. Cornell Vet. 34:312.

Bushnell, L. D., and Payne, L. F.: 1931. Dissemination of pullorum disease in the incubator. Kan. Agr. Exper. Sta., Tech. Bul. 29.

DeOme, K. B.: 1944: The effect of temperature, humidity, and glycol vapor on the viability of air-borne bacteria. Am. Jour. Hyg. 40:239.

———: 1946. Air sanitation. Nulaid News 24:9 (April).

Graham, R., and Michael, V. M.: 1933. Incubator hygiene in the control of pullorum disease. Ill. Agr. Exper. Sta., Cir. 403.

Gwatkin, R.: 1947. Disinfection of incubators with propylene and triethylene glycol. Canad. Jour. Comp. Med. 11:52.

Insko, W. M., Jr., Steele, D. G., and Hinton, C. M.: 1941. Effect of formaldehyde fumigation on the mortality of chick embryos. Ky. Agr. Exper. Sta., Bul. 416:117.

Mohler, J. R.: 1940. Permitted disinfectants. Revised list, 1940. U. S. D. A., Bur. An. Ind., Cir. Letter 2220:1.

Robertson, O. H., Bigg, E., Miller, B. F., and Baker, Z.: 1941. Sterilization of air by certain glycols employed as aerosols. Science 93:213.

Silver, J., Crouch, W. E., and Betts, M. C.: 1942. Rat proofing buildings and premises. U. S. D. I Conservation Bul. 19.

Stafseth, H. J., and Camargo, F.: 1935. On the disinfection of poultry houses by means of fireguns. Jour. Am. Vet. Med. Assn. 86:162.

Storer, T. I., and Mann, M. P.: 1946. Bibliography of Rodent Control. OSRD, Committee on Medical Research, NRC Insect Control Committee Rep. 182:324 + 57 pp. National Research Council, Washington, D. C.

Van Es, L., and Olney, J. F.: 1934. Diseases of poultry—their nature and control. Nebr. Agr. Exper. Sta., Bul. 290.

Yushok, W., and Bear, F. E.: 1944. Poultry manure, its preservation, deodorization and disinfection. N. J. Agr. Exper. Sta., Bul. 707.

# CHAPTER SIX

# PROTEINS, CARBOHYDRATES, FATS, FIBER, MINERALS, AND WATER IN POULTRY FEEDING[1]

*By* L. C. Norris and M. L. Scott, *Department of Poultry Husbandry and School of Nutrition, Cornell University, Ithaca, N. Y.*

☆  ☆  ☆

In order to maintain poultry in good physical condition and to obtain normal growth, egg production, and hatchability, rations must be fed that are adequate in all nutritive essentials. Whenever a serious deficiency of any one of these essential substances occurs, symptoms of the deficiency develop which in many instances are characteristic. These are frequently preceded and accompanied by other symptoms which are nonspecific, such as retarded, uneven growth, rough feather development, decreased egg production, and lowered hatchability. When the deficiency is only a partial one, these may be the only symptoms which are observed. This makes it difficult to recognize the partial deficiency since the nonspecific symptoms may be brought about by a number of causes, including disease. A good background in the nutrition of poultry therefore is necessary for all persons interested in poultry feeding, management, sanitation, and disease.

The nutritive substances of importance in the nutrition and the feeding of poultry are ·classified into groups for purposes of simplification. These groups are (1) proteins and amino acids, (2) carbohydrates, (3) fats, (4) minerals, (5) vitamins, known and unknown, and (6) water. The discussions in this chapter include all of the groups of nutritive substances except the vitamins. These will be discussed in a subsequent chapter.

## THE PROTEINS AND AMINO ACIDS

The proteins are needed by poultry for the synthesis of new body tissue required in growth, to replace body proteins broken down in maintenance, and to furnish the proteins required for egg formation.

*Composition of proteins.* Proteins are complex substances composed of amino acids linked together in chemical combination. The amino acids are composed of carbon, hydrogen, oxygen, nitrogen, and in two instances, sulfur. Approximately twenty different amino acids have been isolated from proteins. The number of amino acids in each protein as well as the percentage

---

ot each is characteristic. Because of this a great many proteins are possible and exist in nature. Every feedstuff contains a number of different proteins.

*Digestion of proteins.* Proteins are not absorbed from the intestinal tract as such but are broken down after ingestion into their constituent amino acids by means of proteolytic enzymes secreted into the gastric, pancreatic, and intestinal fluids. After absorption the amino acids are then rebuilt into characteristic body and egg proteins.

*Amino acid and total protein requirements of poultry for growth and production.* The amino acids found in proteins are given in Table 1. Some

TABLE 1
AMINO ACID REQUIREMENTS OF CHICKS

| Required for Growth and Maintenance | Required Under Certain Conditions | Not Specifically Required |
|---|---|---|
| Arginine | Cystine | Alanine |
| Histidine | Glutamic acid | Aspartic acid |
| Isoleucine | Glycine | Hydroxyproline |
| Leucine | Proline | Norleucine |
| Lysine | Tyrosine | Serine |
| Methionine | | |
| Phenylalanine | | |
| Threonine | | |
| Tryptophane | | |
| Valine | | |

of these amino acids are essential for maintenance and growth of the chick, and therefore must be present in the ration in adequate quantities. A few are synthesized in the body of the chick and may be dispensed with while others either are not required, except when a deficiency of essential amino acids is present in the ration, or are synthesized to some extent, but not in sufficient amounts to promote maximum growth.

The amino acids reported by Almquist (1942a, 1945a, 1945b) and by Almquist and Grau (1944) to be nutritionally essential are given in Column 1, Table 1, those which are essential under certain conditions in Column 2, and those which may be dispensed with are given in Column 3. The amino acid, glycine, in Column 2, is synthesized in sufficient quantities by the chick to promote slow growth, but some must be present in the ration in order to obtain rapid growth. The synthesis of glutamic acid and proline also undoubtedly takes place in the chick, but the evidence at present indicates that this may be limited as is that of glycine.

The amino acid, cystine, is not essential unless the ration is deficient in methionine. When this occurs, the inclusion of protein containing cystine in the ration promotes normal growth. The same situation prevails in the case of tyrosine. When the ration is deficient in phenylalanine, tyrosine becomes an essential amino acid.

Although the nonessential or dispensable amino acids are not nutritionally essential in the ordinary sense, their nitrogen is used by the chick in synthesizing these same amino acids in building body protein. A ration which supplies all of the essential amino acids in just the right proportions and amounts to meet the specific needs for them is not adequate, because it does not contain enough amino acid nitrogen to permit the synthesis of all the protein required for maintenance, rapid growth, and normal egg production. In determining the requirement for each of the essential amino acids for growth and egg production, it is necessary, therefore, to have a sufficient quantity of the dispensable amino acids present in the ration so that the synthesis of these amino acids does not occur at the expense of those which are nutritionally essential.

Although it has been known for many years that the value of proteins for growth and egg production depends upon their amino acid composition, it was impossible to take advantage of this knowledge because of a lack of reasonably complete information concerning the amino acid content of the proteins of feedstuffs, and also a lack of information concerning the quantitative amino acid requirements of poultry. Therefore, in studying the requirements of poultry for protein, extensive studies were made of the supplementary relationships existing between feed proteins. The results of these studies showed that for best results combinations of the mixed proteins of a number of different ingredients were necessary and that with the exception of soybean protein, the inclusion of some protein of animal origin in the over-all combination of proteins was required. Such a combination of proteins was said to be of good quality, meaning that in all probability the protein combination of the feed mixtures contained all of the essential amino acids in the right proportions and amounts for maximum growth or egg production.

Studies on the total protein requirement for growth and egg production have been made in which the basic information on quality of protein combinations for poultry, obtained more or less by trial and error method, has been used. These studies (Heuser, 1941; Hill, 1944) have shown that when the ration contains a protein mixture of good quality, approximately 20 per cent of protein is required in order to promote rapid growth in chicks during the first six to eight weeks of life. Some recent experimental work by Almquist and Asmundson (1944) indicates that a somewhat more rapid rate of growth is obtained by supplying chicks a ration containing approximately 30 per cent protein during the first week or two, and that this effect is not lost during the first eight weeks. However, in recent studies reported by Singsen (1947) it was found that equally good results were obtained in crossbred chicks from diets containing either 21 or 30 per cent protein for the first three weeks and 21 per cent protein for the remainder of the experimental

period of eight weeks. In view of this it appears that the increase in growth obtained by Almquist and Asmundson on the higher protein diet may have been due to additional amounts of the unidentified "animal protein" factor or other growth factors provided by the larger supply of protein supplements used in the high protein ration.

The protein requirement of chicks varies with age. This is due to the fact that in growth much of the protein is required for the formation of new body tissue and little is required for maintenance. The rate of growth of chicks is greatest during the first few weeks after hatching. It gradually declines and becomes zero as the chick attains maturity. Therefore, more protein is required during early chick life than during the last few weeks before cessation of growth.

Although, as previously indicated, the protein requirement during the first six to eight weeks is approximately 20 per cent of the ration, this declines with the rate of growth so that by the time the chick reaches 12 to 14 weeks of age, a ration containing 15 to 16 per cent of protein is adequate. During the later stages of growth, that is from 20 to 24 weeks of age, the protein requirement is probably in the neighborhood of 12 to 14 per cent of the ration. However, it is not practical to feed a ration containing as little protein as this, for any length of time at least. During this stage of growth egg production usually begins, which increases the need for protein.

The results of experimental work on the protein requirements of laying and breeding hens indicate that the ration must contain approximately 15 per cent protein (Heuser, 1941; Hill, 1944). The protein requirement varies somewhat, however, with different strains of hens. Some hens have been reported to give satisfactory egg production on rations containing as little as 13 per cent of protein. The protein requirement of hens also varies obviously with the rate of egg production. When hens are not laying, the protein requirement is probably in the neighborhood of 12 per cent. The protein requirement of the molting hen does not appear to have been studied. However, it is presumably somewhat greater than that of the non-laying hen since the production of a new coat of feathers increases the need for protein.

Since the heavier breeds of chickens grow somewhat faster than White Leghorns, their protein requirements should be somewhat greater, particularly during the earlier stages of growth. The results of an investigation carried out in Australia (Anonymous, 1935) showed that chicks of heavy breeds, such as Light Sussex and Austrolorp, made better gains when a high protein ration was fed to nine weeks of age, while White Leghorns grew equally well when the protein level was reduced at six weeks. Mitchell, Card, and Hamilton (1926, 1931), as a result of studies of growth changes in

chickens, concluded that the protein requirement of White Plymouth Rock chicks was greater than that of White Leghorn chicks.

The results of research work at Cornell and other experiment stations (Heuser, 1946) on the total protein requirement of turkey poults indicate that for rapid growth a ration containing at least 24 per cent of protein of good quality is necessary. Some evidence has been obtained (Fritz, Halpin, and Hooper, 1947), however, that the initial protein requirement of turkeys is higher than this and may be as much as 28 per cent of the ration.

The protein requirement of poults declines with the decreased rate of growth in a manner similar to that of the chick. After six weeks of age the protein level may be lowered to 20 per cent, and after twelve weeks of age, rations containing 16 per cent of protein have been found satisfactory. This appears, however, to be a too rapid rate of decrease in the total protein supplied the poult for maximum growth under all conditions. It seems advisable, therefore, to feed turkey poults a ration containing at least 24 per cent protein during the first eight weeks of life, a ration containing approximately 20 per cent from eight to sixteen weeks, and a ration containing approximately 16 per cent protein from sixteen weeks to market weight.

The protein requirement for breeding turkeys has not been determined experimentally. The results of experience indicate, however, that their requirement is approximately the same as that of laying hens, and that good egg production is maintained by supplying a ration containing 15 per cent protein.

Little is known about the protein requirement of ducks. In the past few years, the age of marketing ducks has been decreased somewhat. This appears to have been brought about by an increase in the amount of protein in the duckling starting ration, and better supplementation with vitamins. At present, some commercial feed manufacturers are supplying a starting ration containing 24 to 25 per cent protein to be fed during the first two weeks, and a growing ration containing about 22 per cent protein to be fed during the next five weeks. After this, a finishing ration containing 16 per cent protein is fed to the market age of approximately nine weeks.

The optimum protein levels for chickens and turkeys, according to the best available information, are given in Table 2. These values were developed by the Subcommittee on Poultry Nutrition of the Committee on Animal Nutrition of the National Research Council, and published by the Council as a report of the Committee on Animal Nutrition. The values are called "nutrient allowances" rather than requirements, since they include necessary margins of safety to take care of differences in requirements due to individual strain and breed variations, and any other variables affecting the requirements.

In formulating poultry rations that contain adequate quantities of protein, it is necessary that all of the essential amino acids be present and in the amounts required to meet all needs for them. The results of studies on the quantitative amino acid requirements of the chick, conducted largely by Almquist and associates, have shown that only arginine, cystine, glycine, lysine, methionine, and tryptophane need to be given particular attention in formulating chick rations. Moreover, it does not appear that special supplementation with glycine-rich protein is necessary except in formulating

TABLE 2

NATIONAL RESEARCH COUNCIL ALLOWANCES FOR PROTEIN AND MINERALS IN POULTRY RATIONS[a]

|  | Starting Chicks 0–8 Weeks | Laying and Breeding Hens | Starting Poults 0–8 Weeks | Breeding Turkeys |
|---|---|---|---|---|
|  | [b] |  | [c] |  |
| Total protein, percentage.... | 20.00 | 15.00 | 24.00 | 15.00 |
| *Minerals:* |  |  |  |  |
| Calcium, percentage....... | 1.00 | 2.25 | 2.00 | 2.25 |
| Phosphorus, percentage[d].... | 0.60 | 0.75 | 1.00 | 0.75 |
| Salt, percentage[e].......... | 0.50 | 0.50 | 0.50 | 0.50 |
| Manganese, mg./lb......... | 25.00 | 15.00 | 25.00 | 15.00 |
| Iodine, mg./lb............. | 0.50 | 0.50 | .. ..... .. | ............ |

[a] Taken from Report No. 1, Nutrient allowances for poultry, Committee on Animal Nutrition, National Research Council, Washington, D. C. (Revised Nov. 1, 1946.)
[b] At eight weeks the total protein can be reduced to 16 per cent.
[c] At eight weeks the total protein can be reduced to 20 per cent.
[d] Inorganic phosphorus should constitute 0.2 per cent of the total feed for chickens, 0.4 per cent of the total feed for turkeys.
[e] This figure represents added salt or sodium chloride.

purified chick rations. All of the other essential amino acids are supplied in sufficient amounts by almost any combination of proteins which may be made, provided the total protein content of the ration is adequate.

The percentages of essential amino acids contained in the more common poultry feedstuffs together with the chick's requirements for these amino acids are presented in Table 3. It is evident from an inspection of the data given in Table 3 that the amino acid composition of the mixed proteins of feedstuffs is not uniform. Soybean oil meal contains more than twice as much lysine and yet only 0.6 as much arginine as peanut meal; corn gluten meal contains much less arginine, cystine, and tryptophane than cottonseed meal or soybean oil meal in spite of the fact that all of these materials have approximately the same total crude protein content.

These differences in the amino acid composition of the proteins of feedstuffs are of great practical significance since they make it necessary to use several different feedstuffs in building poultry rations in order to meet the amino acid requirements without undue wastage of protein. Wastage of

protein must be avoided insofar as possible because of the high cost of protein supplements. The use of excess protein as a means of meeting the need for an essential amino acid not present in sufficient quantities when the total protein is held to the required level is, therefore, highly undesirable.

The need for combining the mixed proteins of several feedstuffs in building a poultry ration in order to meet the amino acid requirements, and the manner in which this is done, is illustrated by Figure 6.1. The figure shows that by combining corn, wheat, soybean meal, fish meal, and alfalfa in proper

TABLE 3

ESSENTIAL AMINO ACID COMPOSITION OF CERTAIN COMMON POULTRY FEED INGREDIENTS

| Ingredient | Crude Protein, Percentage | Amino Acid Composition of Feedstuff[a] (Percentage of Whole Material) | | | | | |
|---|---|---|---|---|---|---|---|
| | | Arginine | Lysine | Methionine | Cystine | Tryptophane | Glycine |
| Alfalfa leaf meal........ | 20 | 0.86 | 0.98 | 0.46 | 0.32 | 0.32 | ........ |
| Barley................. | 10 | 0 48 | 0.18 | 0.21 | 0.18 | 0.08 | ... .... |
| Corn, whole yellow dent.. | 9 | 0.48 | 0.20 | 0.31 | 0.15 | 0.08 | 0.4 |
| Corn gluten meal....... | 43 | 1.7 | 1.1 | 1.0 | 0.47 | 0.26 | 1.7 |
| Cottonseed meal........ | 43 | 3.2 | 1 2 | 0.9 | 0.86 | 0.47 | 2.3 |
| Fish meal...... .. .. | 65 | 3 8 | 3 7 | 1.9 | 0.65 | 0.78 | 2.6 |
| Fish solubles, condensed.. | 35 | 1.5 | 1 7 | 0.52 | 0.21 | 0.14 | ... ..... |
| Meat scrap..... .. | 55 | 3 9 | 2.8 | 1.1 | 0.55 | 0.38 | 2.2 |
| Oats, whole.... · | 10 | 0.58 | 0 33 | 0.23 | 0.15 | 0.12 | ........ |
| Oats, rolled...... | 16 | 0.96 | 0 53 | 0.38 | 0.29 | 0.19 | ........ |
| Peanut meal.... . | 44 | 4.4 | 1 3 | 0.75 | 0.70 | 0.39 | 2.5 |
| Sardine meal... . . | 65 | 4.8 | 3 7 | 2.1 | 0.71 | 0.78 | 2.6 |
| Skimmilk, dried... | 35 | 1 4 | 2 6 | 1.0 | 0.42 | 0.46 | 0.2 |
| Soybean oil meal.. . | 44 | 2 5 | 3 0 | 0 92 | 0.66 | 0.53 | 1.7 |
| Sunflower seed meal | 46 | 3.8 | 2.0 | ¹1.8 | 0.73 | 0.60 | 1.8 |
| Wheat, whole.... | 12 | 0 36 | 0 35 | 0.26 | 0.17 | 0.14 | 0.9 |
| Wheat bran.... ... | 16 | 0 96 | 0.53 | 0 21 | 0.18 | 0.16 | ........ |
| Whey, dried... . . | 12 | 0.43 | 0 84 | 0 34 | 0.36 | 0.24 | 0.0 |
| Yeast, dried brewers'..... | 45 | 1 9 | 3 4 | 0.90 | 0.45 | 0.59 | ........ |
| Chick requirement[b]...... | 20 | 0 9 | 0 9 | 0.5 | 0.3 | 0.2 | 0.8 |

[a] Taken from Almquist (1945a), and Block and Bolling (1945).
[b] Taken from Report No. 1, Nutrient allowances for poultry, Committee on Animal Nutrition, National Research Council, Washington, D. C. (Revised Nov. 1, 1946).

proportions, a ration containing 20 per cent protein is provided in which undue wastage of the essential amino acids of practical importance is avoided.

The figure also shows that it is no longer necessary to depend upon the knowledge of the supplementary relationships of proteins to obtain protein combinations of good quality in poultry rations. The amino acid content of the ration can now be calculated almost as readily as the protein content and compared with the requirement. By proceeding in this manner, greater assurance is obtained that the mixed proteins in the ration are combined in such a way as to give a protein combination of optimum quality.

No work on the amino acid requirements of laying hens has yet been

reported. However, it seems safe to assume that their requirements qualitatively are similar to those of chicks. The proportions of each amino acid required, however, may differ somewhat due to the lower lysine and higher methionine content of egg proteins as compared to body proteins. Furthermore, the amounts needed to meet their requirements are undoubtedly somewhat less than those of rapidly growing chicks in view of the lower total protein requirement.

Extensive studies of the amino acid requirements of turkeys and of ducks
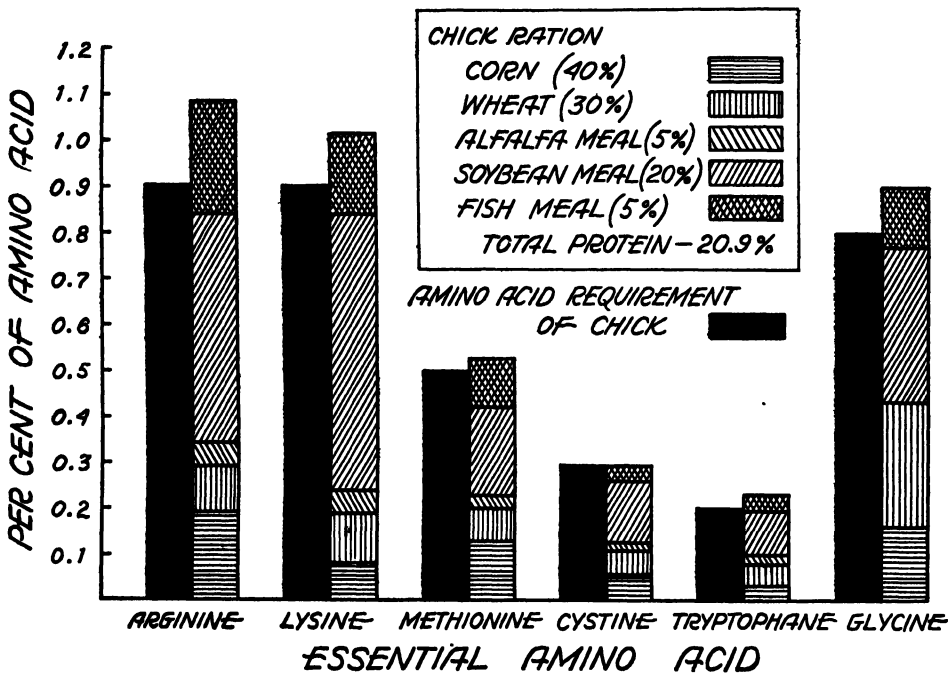


Fig. 6.1. Example showing manner of meeting essential amino acid requirements without undue wastage of protein or amino acids.

have not yet been made. The possibility exists, of course, that their requirements for essential amino acids may be different from those of the chick. For the time being, however, the assumption may be made that the amino acid requirements of these species of domestic fowl are at least qualitatively similar to those of the chick, although the quantitative requirements may be considerably greater, particularly those of poults and ducklings in view of their more rapid rate of growth and higher total protein requirement. In the case of turkeys this assumption is supported by the fact that two groups of investigators (Fritz, Hooper, Halpin, and Moore, 1946; Grau, Kratzer, and

Asmundson, 1946) have found the poult's requirement for the amino acid, lysine, to be approximately 1.3 times that of the chick.

*Other functions of amino acids.* Amino acids fulfill many other functions aside from their main action as building stones for the new tissues required in growth and maintenance. Creatine, which is essential physiologically for the functioning of muscular tissue, is synthesized in the body from arginine and glycine. The amino acid, tyrosine, is used in the formation of the hormone, thyroxine, and is necessary for the formation of certain



Fig. 6.2. Lack of normal feather pigmentation in a bronze poult caused by a deficiency of lysine.

pigments in the feathers of colored fowl. Recently lysine has been shown to be needed in some indirect way for feather pigment formation in turkeys. The feather achroma obtained by Fritz and associates (1946) in bronze turkeys fed a corn gluten meal diet was found to be corrected by the addition of lysine. The picture in Figure 6.2 shows the abnormal white wing feathers of bronze poults obtained when the diet was deficient in lysine.

Indirect evidence (McGinnis, Norris, and Heuser, 1944a) indicates also that methionine is needed for the formation of choline in mature chicks and probably also in growing chicks when other necessary precursors of choline are present in the ration. Evidence has been obtained (Rosen, Huff, and Perlzweig, 1946) that the amino acid, tryptophane, is either converted into

nicotinic acid or stimulates its synthesis. Thus, when the protein of a ration is deficient in tryptophane, the requirement for niacin is increased.

*Effect of heat upon protein quality.* Experimental results have shown that moderate heat treatment greatly improves the quality of soybean proteins for poultry. The poor quality of raw soybeans is apparently due to an unknown substance in the bean which counteracts the action of the enzyme, trypsin, in the intestinal tract, and thereby reduces the digestibility of the proteins (Ham, Sandstedt, and Mussehl, 1945; McGinnis and Menzies, 1946). The unknown substance has been found to reduce the digestibility of other proteins besides those present in soybeans.

Prolonged heat treatment at high temperatures, on the other hand, decreases the quality of proteins including those of soybeans. Experimental work (Greaves and Morgan, 1934) has shown that the amino acids which are affected by heat treatment are lysine and histidine. The lysine is apparently not destroyed by the heat treatment since analysis for lysine content has indicated no material change (Block, Jones, and Gersdorff, 1934). These results suggest that the lysine has been tied up with some other substance in such a way that it is not freed during the digestion of the heated proteins, and hence does not become available. Whether or not histidine is tied up in a similar manner or is actually destroyed does not appear to have been determined.

*Sources of protein.* The feedstuffs which are commonly used as protein supplements in poultry rations at the present time are soybean meal, meat scraps, and fish meal. Peanut meal and corn gluten meal are used to a limited extent, but are not important sources of protein for feeding poultry. With the development of a method for detoxifying cottonseed meal (Boatner and Hall, 1946; Groschke, Rubin, and Bird, 1947), it is possible that this product may be used much more extensively as a source of protein. However, none of the vegetable protein supplements, except soybean meal, contain protein of approximately the same composition of essential amino acids as contained in meat scrap and fish meal. Hence the vegetable proteins are not particularly adapted in general for poultry feeding.

An inspection of the amino acid composition of the other vegetable protein supplements, given in Table 3, shows that their chief deficiency is in the amino acid, lysine, of which large quantities are required for growth and the formation of egg proteins. Evidence has been obtained (Heuser, Norris, and McGinnis, 1946), however, that satisfactory growth can be obtained by combining corn gluten meal, cottonseed meal, and peanut meal in limited quantities with soybean meal and with meat scraps and fish meal. The quantities of these vegetable protein supplements included in the ration must be restricted, however, in order to meet the requirements for lysine.

Dried skimmilk and dried buttermilk are also excellent sources of protein of superior quality. Their protein content, however, is lower than that of

the protein supplements just discussed. When used in poultry rations, they are used more for their content of essential vitamins than for proteins because of their relatively high cost. At present, little dried skimmilk and dried buttermilk are available for poultry feeding because of the demand for human use. Whether or not this demand will change with improved world conditions is problematical.

*Influence of protein level upon egg production.* The protein level of the ration fed to chicks and growing pullets appears to have little effect on the age at which the pullets begin to lay or upon subsequent egg production except when it is low enough to retard growth greatly. Carver and associates (1932), however, reported that a protein level of 12 per cent retarded the age of sexual maturity of White Leghorn pullets from 25 to 40 days, but subsequent results of other workers on the riboflavin requirements showed that the low-protein ration was greatly deficient in this vitamin. Later Carver and associates (1939) reported that pullets fed rations containing 19 per cent protein reached sexual maturity a few days earlier than those fed rations containing 13 per cent. Several other groups of investigators (Winter, Dakan, and Bayes, 1932; Morris, Thompson, and Heller, 1932; Heuser and Norris, 1933; Byerly, Titus, and Ellis, 1933; Tepper, Durgin, and Charles, 1939) have obtained results indicating that the rate of sexual maturity was not influenced to any marked degree by the protein content of the ration.

Carver and associates (1939) fed protein levels varying from 13 to 19 per cent during the first twenty-two weeks after hatching and 15.3 per cent thereafter, and found during the first 224 days of production that neither the egg weight nor the rate of egg production was influenced by the quantity of protein fed during the growing period. Bronkhorst (1938) also found that egg yield was not affected by the amount of protein fed growing pullets during the pre-laying period.

Hens in active egg production require an adequate continuous supply of protein since the nitrogen required for egg production must come from the feed (Wilcox, 1934). Insufficient protein in the ration of laying hens will result in decreased egg production, lower body weight, and smaller egg size. Heuser (1936) found that a ration containing 12 per cent protein was insufficient to maintain either satisfactory egg production or egg size, while a ration containing 14 per cent gave satisfactory egg production, but did not maintain body weight at all times and was not conducive to best egg size. Satisfactory maintenance of weight and satisfactory egg size, however, were obtained when the ration contained 15 to 16 per cent protein. Heuser's observations have been confirmed by Byerly, Titus, and Ellis (1933) and by Heiman and associates (1936).

*Protein level and feathering.* Because feathers are composed chiefly of protein, it is obvious that poor feathering will result from the lack of ade-

quate protein in the ration. Tomhave (1939) has shown that when the protein level in the ration was less than 18 per cent during the first eight weeks, bare breasts occurred in White Leghorn pullets. Several other investigators (Gericke and Platt, 1932; McConachie, Graham, and Branion, 1935; Ackerson, Blish, and Mussehl, 1939) have reported that feather development improved as the protein level in the ration was increased.

A possible relationship between the protein content of the ration and feather pulling, tail picking, and cannibalism has been reported by Margolf (1929). In his experiments, these vices developed in chicks on low protein rations as early as the second and third weeks.

*Excess protein.* The question as to whether the feeding of excess protein to chickens is harmful remains unsettled. McConachie and associates (1935) and Milne (1932) have reported detrimental effects upon growth rates when the protein level of the ration was raised to 30 per cent or over, and the former found a high mortality of chicks at a 35 per cent level. Whether or not the retarded growth and increased mortality were due to the ingestion of excess protein directly, or indirectly as a consequence of a lack of balance of other nutrients created by including large amounts of high protein concentrates in the ration, cannot be determined from the evidence, especially since Almquist and Asmundson (1944) have obtained improved growth of chicks on a 30 per cent protein ration. Heuser (unpublished results, Cornell University) has obtained normal results with hens fed all-mash diets containing as much as 28 per cent protein over prolonged periods.

*Uremic poisoning.* Patterson (1928) has suggested that the nutritional gout or uremic poisoning in chickens, other than that caused by vitamin A deficiency, may be due to the feeding of excess nitrogenous concentrates. This condition, which is characterized by internal deposits of sodium urate, particularly in the kidneys and ureters, has been observed also by other workers (Mayall, 1929; Hartwick, 1940; Bird, Rubin, Whitson, and Haynes, 1946). Each of these workers appears to have a different explanation for the cause of this condition.

In support of Patterson's suggestion, Schlotthauer and Bollman (1934) have reported that they were able to produce gout in turkeys by increasing the protein level of the turkey diet to 40 per cent by the addition of horse meat to a standard turkey ration. They were able to produce this effect also by the addition of 5 per cent urea to the diet. On the other hand, Patton (1939) has reported that hens are able to tolerate large single doses of urea.

The injection of large amounts of glycine and of *dl*-alanine have been shown by Patton (1939) to be toxic when given to White Leghorn hens. Upon autopsy, the hens fed excess glycine showed greatly enlarged kidneys which upon histological examination revealed indications of incipient necrosis. According to Patton, the evidence showed conclusively that the

kidneys were the chief site of glycine toxicity. In spite of the fact that the hens died from this treatment, Patton did not report an accumulation of sodium urate in the tubules of the kidneys.

Bird and associates (1946) found that a large percentage of the newly hatched chicks from hens receiving a diet deficient in animal protein consistently showed urate deposits in the kidneys and ureters. This condition could be prevented by including sardine meal or cow manure in the breeding diet fed the hens. Thus, it appears that under normal conditions nutritional gout in poultry may be due to the deficiency of an unknown vitamin.

## CARBOHYDRATES

Carbohydrates, along with fats, provide the energy needed by poultry for growth, maintenance, and reproduction. Carbohydrates, however, play a much more important role than fats in providing energy because they constitute a much larger proportion of the ration.

*The nature of carbohydrates.* Sugars, starches, dextrins, pentosans, and celluloses are the chief members of a group of organic compounds which are referred to as carbohydrates. All carbohydrates are composed of carbon, hydrogen, and oxygen, the latter two elements always being present in the ratio of two atoms of hydrogen to one atom of oxygen.

The sugars are the structural units from which all carbohydrates are formed. The simple six-carbon sugars are known as monosaccharides; the more complex sugars, such as sucrose, maltose, and lactose, are known as disaccharides, while the starches and celluloses are composed of many glucose molecules and are therefore termed polysaccharides.

*Digestion of carbohydrates.* Only the simple sugars have sufficiently small molecular structures to gain entrance into the blood stream from the intestines. Therefore, all carbohydrates must be broken down into their simplest constituents before they can be used in animal metabolism. The animal tissues secrete certain enzymes, amylases, which are capable of splitting starches and dextrins into the disaccharide, maltose. Another enzyme, maltase, is then capable of breaking maltose down to glucose, in which form the carbohydrate may be absorbed into the blood stream. Other specific enzymes in the digestive tract act upon the other complex carbohydrates, breaking them down into their structural units, the simple sugars. Following absorption into the portal vein, the simple sugars are carried to the liver where they are converted into glycogen. Then, as the demand arises in the body for energy, glycogen breaks down, releasing glucose to be carried by the blood to the site showing the demand. Thus glucose performs the major role in carbohydrate metabolism. When the glycogen stores become filled, the excess sugar, resulting from carbohydrate digestion, is readily converted into fat and is stored as such in the various fat depots throughout the animal body.

Cellulose and an accompanying compound, lignin, which together make up the cell wall structure of plants, are not acted upon by any enzyme secreted by animal tissues, but can be broken down by certain bacteria (Maynard, 1947). Although lignin is not a carbohydrate, it is discussed along with carbohydrates because it occurs in intimate association with cellulose. An excellent review of the present knowledge regarding lignin has been made by Hibbert (1942).

Unlike the ruminant which fosters a host of cellulose-splitting microorganisms within its rumen, the chicken is almost totally unable to derive any benefit from cellulose. The enzymes of the digestive tract of the chicken appear to be as efficient as those of any other animal in breaking down starch and dextrins, provided, of course, that these nutrients are not enveloped by a cellulose membrane which protects them from the action of the digestive juices. Such a condition as this does appear to exist in certain feedstuffs such as wheat bran, thereby greatly lowering the available energy content of these feeds as compared with other feeds containing equally as much cellulose, but having the starch and sugar fractions accessible to digestion.

*Chemical determination of carbohydrates.* About eighty years ago, Henneberg and Stohmann (1860–65) of the Weende Experiment Station in Germany, realized that in order to formulate rations which contained adequate energy they needed a chemical method for determining the indigestible portion of the carbohydrate fraction. The method which they devised is the one still used in most laboratories for determining the crude fiber content of a feed. In brief, the method entails digestion of the feedstuff first in dilute acid, then in dilute alkali, followed by a determination of the percentage of the feed remaining undissolved.

Since some of the pentosans and lignins which are totally indigestible by the chicken are slightly soluble in dilute acids and alkalies, this method does not give an absolute indication of digestibility, but it is useful because, on the whole, digestibility correlates rather well as an inverse function of the crude fiber content of the feed material.

In the chemical analysis of feeds, the carbohydrate portion is divided into two classifications, the crude fiber representing the indigestible portion, and the nitrogen-free extract, the supposedly digestible portion. The nitrogen-free extract contains the sugars and starch, but as has been pointed out, it also contains some indigestible pentosans and lignins. It is determined by taking the sum of the moisture, protein, fat, ash, and fiber of a feed and subtracting this figure from 100. It is clear, in view of the many possibilities for variability in composition of the nitrogen-free extract, that this value cannot be relied upon to serve as an index of the carbohydrate content of a feed available for energy purposes.

Consequently, the best method of determining the energy content of a feedstuff is by feeding it and determining the calories derived by the chick

for gain and maintenance from a given quantity of feed. This has been done with a wide variety of materials by Fraps (1946). A discussion of his findings will be presented under the section on energy.

*Cereals and cereal by-products.* The cereal grains are universally recognized as our best sources of available, energy-producing carbohydrate. The four most widely used cereals, corn, wheat, barley, and oats, appear to rank in the order mentioned as sources of this nutrient. Experience has shown that the first two cereals can be interchanged at will, provided, of course, that the remainder of the diet is adequate in all nutrients other than carbohydrate. Oats and barley contain too much fiber to be used effectively as the sole source of carbohydrate. Removal of the oat hull, as is done in the manufacture of feeding rolled oats, results in a product having an energy content equal to that of corn or wheat.

The by-products resulting from processing of the cereal grains are much inferior to the whole grains as sources of carbohydrate. This is due to the fact that the outer coating of the cereal grains, which is contained in practically all of the by-products, is high in fiber and therefore low in digestibility. Fraps (1946) has found, indeed, that oat hulls contain no available energy for the chick.

*Milk and milk by-products.* Since the most important available carbohydrate in feedstuffs is starch, most feedstuffs can be evaluated as a source of carbohydrate by determining the starch content. An exception to this is found in the case of milk and milk by-products where the carbohydrate is present in the form of the disaccharide, lactose. While the digestive tract of the chicken contains an enzyme or enzymes for the splitting of lactose into its constituent monosaccharides, glucose and galactose, this hydrolysis appears to progress at a fairly slow rate.

Experience has shown that while small amounts, up to 10 per cent, of dried milk by-products have a definite beneficial effect upon the growth of chicks, raising the level of lactose in the diet too high results in retardation of growth. This effect is quite probably the result of two phenomena. First, the slow rate of hydrolysis of lactose results in a lowering of the sugar uptake by the blood stream and therefore a lowering of the available energy for growth. In addition, the presence of large amounts of unhydrolyzed lactose in the ceca and lower intestines presents an excellent carbohydrate source for the growth of acidophilic microorganisms (Hull and Rettger, 1917). The tremendous multiplication of these microorganisms results in abnormal digestion in the lower gut and produces a severe diarrhea which flushes out many of the nutrients which might otherwise be absorbed from this site.

## FATS

Fats, like carbohydrates, are composed of carbon, hydrogen, and oxygen, and are used by the body as a source of energy. Since fats contain more

carbon and hydrogen and less oxygen than do carbohydrates, they contain about 2.25 times as much energy per unit weight.

*Digestion of fat.* After ingestion, fats are broken down by enzymes present in the intestinal juices into their constituent parts, the fatty acids and glycerol, in order to be absorbed through the intestinal wall. These constituents following absorption are reunited into fat which is carried to all parts of the body by way of the lymph and blood channels. Much of this fat is probably used at once as energy. Any excess, however, is deposited within the cells of the body and in the fat depots underneath the skin, in the abdominal cavity, and around certain vital organs. Conclusive evidence that fatty acids are first converted into carbohydrates before being used to supply energy is still lacking, but glycerol may be converted into glucose and stored temporarily in the liver and muscle tissues as glycogen.

In fattening poultry, it is much more feasible to use a ration high in carbohydrates than one high in fat for several reasons. Fats are digested more slowly than are carbohydrates. A high level of fat in the ration tends to retard digestion of other nutrients. Fats are much more expensive than carbohydrates, and the tendency during recent years has been to remove more and more of the fat from the ingredients used in poultry rations since these fats demand a much higher price in other markets.

*Role of fat in poultry nutrition.* Fat has other functions in nutrition in addition to serving as a source of energy. Burr and Burr (1930) have shown that the rat requires certain unsaturated fatty acids as essential nutrients in the ration. Thus far no evidence has been produced to show that poultry also have this requirement. Indeed, Russell (1939) has reported that the growing chick can do very well on rations containing as little as one-tenth of 1 per cent fat.

On the other hand, some fat in the diet is highly desirable since it acts as a carrier and aids in the absorption of the all-important fat-soluble vitamins (Russell, Taylor, Walker, and Polskin, 1942). In addition to this, its high energy content many times has served to provide the necessary energy in rations which would otherwise be inadequate due to a high percentage of fiber. The fat level which appears at present to be economically feasibly is in the neighborhood of three to four per cent of the poultry ration.

*Importance of antioxidants.* The character of the fat is of extreme importance in poultry feeding. Certain fatty acids, when not protected adequately by natural antioxidants, may combine with oxygen from the air, forming organic peroxides. These peroxides in turn may act to destroy the vitamin E and vitamin A activity of the feed (Mattill, 1927; Smith, 1939), thereby producing a deficiency of these vitamins even though they were present in ample amounts at the time the feed left the manufacturer. Thus,

it is of importance in the formulation of a feed to use ingredients which supply fat free of "rancidity," and if possible, containing antioxidant properties. Vitamin E acts as an effective antioxidant, but in the process becomes oxidized itself. Most of the natural carriers of fat contain unknown antioxidants which are sufficient to protect the fat for a considerable period of time, provided they have not been destroyed by heat or lost in some other way during the processing of the material.

## ENERGY

Of the various nutrients needed for growth and production, the requirement for energy is by far the largest and primarily governs the total food intake. Yet, in most poultry feeding, major attention has been given to amino acids, minerals, and vitamins with a tendency to overlook the energy requirement. This has been due, in part, to the fact that a deficiency of energy in a poultry ration usually does not express itself in terms of definite symptoms other than in general lowered growth and reduced efficiency of feed utilization. Therefore, since no spectacular diseases have resulted even when a ration was seriously deficient in energy, the study of this nutrient has been neglected in favor of the study of protein, mineral, and vitamin deficiencies where, at times, even a moderate deficiency may result in lesions which are easily discerned.

The results of studies recently reported by Scott and associates (1947) of the Storrs Agricultural Experiment Station, show that best growth of chicks was obtained when the ration contained 68 per cent of corn meal and a low percentage of fiber. Growth, feathering, and efficiency of feed utilization were impaired in direct proportion to the lowering of the available energy content of the ration, as for example, by substitution of pulverized oats for a part of the corn meal.

In studies with turkey poults conducted at Cornell by Scott and Heuser, the results of which have not yet been published, it has been found that increasing the available energy content of the poult ration resulted in a marked increase in growth. This was accomplished by the substitution of corn meal, ground whole wheat, and feeding rolled oats for the wheat bran, wheat middlings, pulverized oats, and alfalfa meal in a turkey starter ration. The substitutions resulted in increases in poult growth of approximately 100 grams at four weeks of age.

Results such as these serve to emphasize the importance of carefully considering energy content when formulating rations for poultry. In order to do this, however, it is necessary to have adequate data as to the available energy content of all of the feedstuffs to be used in making up the ration.

A comprehensive investigation of the energy values of poultry feedstuffs

## TABLE 4

**AVERAGE COMPOSITION AND APPROXIMATE AVAILABLE ENERGY CONTENT OF VARIOUS MATERIALS USED IN POULTRY FEEDING[a]**

| Material | Mineral Matter Percentage | Calcium Percentage | Phosphorus Percentage | Manganese mg./lb. | Protein Percentage | Fat Percentage | Nitrogen-free Extract Percentage | Fiber Percentage | Available Energy Cal./lb. |
|---|---|---|---|---|---|---|---|---|---|
| Alfalfa leaf meal | 12.2 | 1.90 | 0.22 | 11.8 | 21.1 | 2.8 | 39.8 | 16.1 | 314 |
| Barley, whole | 2.9 | 0.05 | 0.38 | 6.4 | 11.8 | 2.0 | 68.0 | 5.7 | 811 |
| Blood meal | 3.8 | 0.33 | 0.26 | 0.6 | 82.2 | 1.2 | 2.7 | 1.3 | 1004 |
| Bone meal, steamed | 81.3 | 32.61 | 15.17 | 5.9 | 7.1 | 3.3 | 3.9 | 0.8 | 202 |
| Brewers' grains, dried | 3.7 | 0.25 | 0.47 | 8.6 | 25.6 | 6.7 | 42.0 | 14.8 | 1005 |
| Buttermilk, dried | 10.5 | 1.36 | 0.74 | 0.2[b] | 33.8 | 5.6 | 41.9 | 0.4 | 707 |
| Casein | 3.9 | | | 1.1 | 81.9 | 0.1 | 3.3 | 0.2 | 1016 |
| Corn, whole No. 2 | 1.3 | 0.01 | 0.27 | 2.2 | 9.4 | 3.9 | 68.4 | 2.2 | 1145 |
| Corn bran | 2.3 | 0.03 | 0.27 | 7.3 | 9.8 | 6.4 | 61.8 | 9.8 | 562 |
| Corn distillers dried solubles | 2.9 | 0.05 | 0.31 | 11.0 | 30.6 | 10.6 | 38.7 | 10.8 | 853 |
| Corn gluten feed | 6.1 | 0.14 | 0.55 | 12.0 | 26.4 | 2.5 | 48.4 | 7.1 | 565 |
| Corn gluten meal | 1.8 | 0.03 | 0.38 | 1.8 | 42.2 | 2.3 | 42.0 | 2.5 | 839 |
| Cottonseed meal | 5.5 | 0.24 | 1.11 | 8.2 | 43.2 | 7.2 | 27.0 | 10.6 | 694 |
| Fish meal | 14.7 | 4.24 | 3.06 | 18.7 | 66.7 | 6.8 | 3.6 | 0.7 | 898 |
| Kafir grain | 1.7 | 0.04 | 0.30 | | 11.2 | 3.0 | 70.3 | 2.3 | 1078 |
| Liver and glandular meal | 5.0 | 0.70 | 0.90 | 1.9 | 65.0 | 12.0 | 3.2 | 1.5 | 1092 |
| Meat and bone scraps | 24.6 | 8.70 | 4.30 | 8.2 | 55.0 | 10.7 | 1.2 | 2.2 | 724 |
| Milk, dried skim | 8.0 | 1.24 | 0.96 | 0.2 | 34.8 | 0.9 | 50.1 | 0.0 | 525 |
| Oats, whole | 3.6 | 0.09 | 0.33 | 16.4 | 12.0 | 4.7 | 60.2 | 10.6 | 760 |
| Oat meal, feeding | 2.4 | 0 06 | 0.45 | 16.4 | 16.3 | 5.9 | 64.1 | 2.8 | 1155 |
| Oat hulls | 6.6 | | | | 4.2 | 1.6 | 49.9 | 31.5 | 0 |
| Peanut meal | 6.3 | 0.17 | 0.55 | | 42.7 | 8.5 | 27.0 | 8.9 | 731 |
| Sardine meal | 15.2 | 4.4[b] | 3.2[b] | 19.0[b] | 64.5 | 9.8 | 3.8 | 0.2 | 825 |
| Sesame meal | 12.0 | 2.02 | 1.61 | | 39.6 | 12.6 | 23.2 | 6.1 | 706 |
| Soybeans | 5.3 | 0.20 | 0.60 | 14.5 | 36.9 | 17.2 | 26.3 | 4.5 | 1070 |
| Soybean meal, expeller | 5.7 | 0.28 | 0.66 | 13.6 | 44.3 | 5.7 | 30.3 | 5.6 | 674 |
| Soybean meal, solvent | 6.0 | 0.29 | 0.68 | 13.6 | 46.4 | 1.6 | 31.7 | 5.9 | 565 |
| Sunflower seed oil cake | 4.2 | 0.43 | 1.04 | | 34.8 | 18.3 | 21.8 | 10.9 | 1100 |
| Wheat, whole | 2.0 | 0.03 | 0 43 | 14.1 | 13.1 | 1.7 | 70.0 | 3.0 | 1024 |
| Wheat, bran | 6.0 | 0.12 | 1.32 | 49.2 | 15.8 | 5.0 | 54.3 | 9.5 | 478 |
| Wheat brown shorts | 4.4 | | | 45.5 | 17.8 | 4.7 | 57.0 | 6.2 | 581 |
| Wheat flour middlings | 3.4 | 0.09 | 0.72 | 28.7 | 17.0 | 4.9 | 59.9 | 4.4 | 720 |
| Wheat, red dog | 2.6 | 0.07 | 0.52 | 15.9 | 16.9 | 4.0 | 63.3 | 2.4 | 1020 |
| Whey, dried | 9.7 | 1.18 | 0.66 | 6.4 | 12.5 | 0.7 | 72.1 | 0.0 | 490 |
| Yeast, dried brewers' | 7.0 | 1.48 | 1.28 | 1.1 | 45.0 | 3.0 | 36.0 | 1.0 | 476 |

has been made by Fraps (1946) of the Division of Chemistry of the Texas Agricultural Experiment Station. His results, presented in the last column of Table 4, make it possible to formulate a ration of any desired energy content.

From an inspection of the data given in Table 4, it is evident that the available energy content of a feedstuff depends upon several different factors. In general, as the fiber content goes up the energy content is correspondingly lowered, but this is not true in all cases. For example, sunflower seed oil cake contains the same energy content as corn meal, and yet its fiber content is much greater. This undoubtedly is due to the very high ether extract or fat content of sunflower seed oil cake. On the other hand, dried brewers' yeast, containing less fiber than corn meal, also contains much less available energy. This is probably due to the nature of the nitrogen-free extract of the yeast. The fact that proteins can be used as energy is apparent from the energy values given for the animal products. Fish meal, having a higher percentage of protein, contains more energy than meat and bone scraps, even though the latter material contains a higher level of fat.

Fraps has calculated the average energy content of a number of poultry rations recommended by various experts in poultry nutrition. The average energy in Calories per pound, was found to be 816 for all-mash chick starter, 879 for all-mash growing ration, and 917 for mash and grain; 831 for all-mash laying ration, 895 for laying mash with grain; 814 for all-mash breeding ration and 865 for breeding mash with grain. However, a calculation of the energy content of the corn meal ration found by Scott and associates (1947) to give best growth of chick broilers shows that this ration contained about 1,000 Calories per pound, indicating that for best results rations of somewhat higher energy content than have been used in the past may be desirable, particularly for very young stock.

The availability of data concerning energy content, as well as amino acid, mineral, and vitamin content of poultry feedstuffs, together with fairly accurate information regarding the requirements of poultry for these nutrients, makes it possible to formulate rations which theoretically contain all of the nutrients essential for optimum growth and production. On the other hand, recommendations of mixtures for poultry feeding should always be put to the test of practical experiment since quality of feeds varies to a considerable extent, and oftentimes the quality of the animal produced cannot be predicted from an inspection of the ration fed.

## MINERAL ELEMENTS

Mineral elements in properly balanced amounts are as important in the maintenance of the life, well-being, and production of poultry as amino acids and vitamins. They enter into the composition of the bones and give the skeleton, the bony framework of the body, the rigidity and strength needed

to support the soft tissues. Minerals combine with protein, lipids, and other substances which make up the soft tissues of the body. They take part in the maintenance of osmotic pressure and the acid-base balance, and exert specific effects on the ability of muscles and nerves to respond to stimuli. Minerals are also necessary for the activity of some of the enzymes present in the body.

*Essential minerals.* The mineral elements which have been found essential for the maintenance of animal life in general are calcium, phosphorus, magnesium, sodium, potasium, chlorine, manganese, iodine, iron, copper, sulfur, and zinc. Cobalt appears to be an essential element for ruminants, but no evidence has been obtained that it is necessary for poultry or other types of farm animals. Fluorine in small amounts is a constant constituent of several tissues, particularly bones. Evidence has been obtained that

TABLE 5
MINERAL COMPOSITION OF MATURE FOWLS, FAT FREE, DRY BASIS

|  | Cockerel | | Pullet | |
|---|---|---|---|---|
|  | Flesh and Offal | Bones | Flesh and Offal | Bones |
| Ash............... | 5.50% | 38.88% | 4.98% | 40.50% |
| Ash............... | 17.640 gm. | 68.740 gm. | 14.220 gm. | 55.890 gm. |
| Phosphorus......... | 2.848 " | 11.600 " | 2.418 " | 9.880 " |
| Sulfur............. | 3.733 " | 0.922 " | 3.160 " | 0.742 " |
| Calcium........... | 0.430 " | 25.680 " | 0.345 " | 20.770 " |
| Magnesium......... | 0.478 " | 0.807 " | 0.374 " | 0.624 " |
| Potassium.......... | 4.778 " | 0.787 " | 3.630 " | 0.566 " |
| Sodium............ | 1.443 " | 0.713 " | 1.113 " | 0.595 " |
| Chlorine........... | 2.283 " | 0.817 " | 1.996 " | 0.552 " |
| Iron............... | 0.131 " | 0.030 " | 0.128 " | 0.034 " |

traces of this element may be essential, or at least beneficial, for some species, but no direct evidence to this effect has been obtained with poultry.

*Mineral content of tissues and eggs.* The distribution of minerals in the body of mature chickens is illustrated by the work of Halnan (1936), who made a detailed mineral analysis of a Light Sussex cockerel and pullet. The results of the analysis are presented in Table 5. They show that approximately 80 per cent of the total ash is present in the bone. The analyses of the individual mineral constituents show, furthermore, that the major portion of the calcium, magnesium, and phosphorus of the body is present in the bones. Chlorine, iron, sodium, sulfur, and potassium, on the other hand, are largely present in muscles and other soft tissues.

Spectrographic analyses of eggs and chick tissues have been carried out by Drea (1935). The elements which pass from the feed into the hen's blood, from there into the egg, and finally into the chick's tissues and blood were aluminum, barium, calcium, copper, iron, magnesium, phosphorus, potas-

sium, rubidium, sodium, silicon, strontium, titanium, and vanadium. Manganese and zinc, although present in the hen's ration, were absent from the chick's blood, but present in all organs but one. Boron and silver were present in the egg, but fluorine was found only in one femur. Chromium, lead, and molybdenum were found in the hen's blood, but were not constantly present in the eggs.

The presence in the body tissues of those mineral elements not found essential is probably in most instances fortuitous, and is due to the fact that they are universally present in the food supply. Silicon, however, is the chief constituent of the ash of feathers, and may possibly exercise a role in maintaining their rigidity.

*Minerals in feedstuffs.* The minerals of greatest importance in the growth of chicks from the quantitative point of view are calcium, chlorine, magnesium, phosphorus, potassium, and sodium. A consideration of these and other mineral requirements shows that many feedstuffs are deficient in minerals. The feedstuffs of plant origin are low in chlorine and sodium, and with the exception of alfalfa meal, also low in calcium. They contain relatively large amounts of potassium and reasonable quantities of magnesium and phosphorus. Phosphorus, however, is largely in the form of phytin, which according to recent evidence is not made available in any quantity by the digestive processes of the chick. Corn and the feedstuffs of animal origin are low in manganese. With the exception of manganese and, in the case of milk products, also iron and copper, the feedstuffs of animal origin are reasonably well supplied with minerals. It is necessary, therefore, to supplement poultry rations with additional sodium and chlorine in the form of salt, additional calcium, and, in many instances, phosphorus and manganese. Because of the use of large amounts of soybean meal in present-day poultry rations, supplementation with additional iodine is usually indicated, unless large quantities of fish by-products are included in the ration, owing to the fact that soybeans have goitrogenic properties (Wilgus, Gassner, Patton, and Gustavson, 1941a). This is usually added by the use of iodized salt. Since salt is added routinely, the only minerals generally given special consideration in formulating poultry rations are calcium, phosphorus, and manganese. The amounts of these minerals present in the commonly used poultry feedstuffs are presented in Table 4.

*Calcium and phosphorus.* Calcium and phosphorus are discussed together because of their close association in metabolism, particularly in the formation of bone. According to the work of Halnan (1936) about 98.3 per cent of the total calcium in the body of the mature fowl is present in the bones, and the remainder in the soft tissues. The proportion of phosphorus present in the bones of the mature fowl, on the other hand, is somewhat less than that of calcium, amounting to approximately 80 per cent of the total phosphorus

in the body, the other 20 per cent being required for various functions throughout the body.

In the growing chicken, the major portion of the calcium in the ration is used for bone formation, while in the mature fowl the major portion is used for egg shell formation. Calcium is also essential for clotting of the blood, is required, along with sodium and potassium, for the normal beating of the heart, and is concerned in the maintenance of acid-base equilibrium.

In addition to its role in bone formation, phosphorus exercises important functions in the metabolism of carbohydrates and fats, it enters into the composition of important constituents of all living cells, and salts formed from it play an important part in the maintenance of the acid-base balance. It is apparently also concerned in calcium transport in egg formation.

The utilization of calcium and phosphorus is dependent upon the presence of an adequate amount of vitamin D in the ration. In the absence of sufficient quantities of this vitamin, calcium and phosphorus are not deposited in a normal manner in the bones of growing chicks, or if the deficiency is greatly prolonged these elements may even be withdrawn from the bones. This disturbance in calcium and phosphorus metabolism results in a severe lameness called rickets. In vitamin D deficiency these minerals are also withdrawn from the bones of mature fowls. Unless the vitamin D deficiency is accompanied by a marked deficiency of phosphorus, the blood calcium level is reduced in avian rickets rather than the blood phosphorus level.

The quantity of calcium and phosphorus required by poultry is dependent to some extent upon the level of vitamin D supplied in the ration. When large amounts of vitamin D are fed, the quantities of calcium and phosphorus in the ration may be reduced. On the other hand, a deficiency of vitamin D can be offset to a considerable extent by increasing the quantities of calcium and phosphorus.

The ratio of calcium to phosphorus must be considered in formulating rations in view of their close relationship in metabolism. The most desirable ratio of calcium to phosphorus depends upon several factors and may be varied over a fairly wide range without serious harm. In the case of the growing chick it is now generally accepted that the most desirable ratio lies between 1.5:1 and 2:1. For the laying hen the ratio is considerably wider due to the higher requirement of the hen for calcium. When either element is present in large excess it interferes with the absorption of the other from the digestive tract.

All of the common sources of calcium used in feeding poultry appear to be well utilized. On the other hand, some of the sources of phosphorus are not readily utilized by poultry, and the utilization is dependent, in part, upon the character of the vitamin D included in the ration. McGinnis, Norris, and

Heuser (1944b) have shown that more than sixteen times as much vitamin D is required to promote normal bone development in chicks when the phosphorus is supplied entirely by means of feedstuffs of vegetable origin than when supplied in equivalent amounts in a purified diet by means of inorganic salts. The phosphorus in the feedstuffs of vegetable origin is largely in the form of phytin which, when fed as the only source of phosphorus in a purified diet, has been found by Gillis, Norris, and Heuser (unpublished results, Cornell University), to provide no available phosphorus, although the vitamin D content of the diet was in excess of the requirement under ordinary conditions. Singsen, Matterson, and Scott (1947) have shown that pure vitamin $D_3$ is more effective in making the phosphorus of phytin available than is the vitamin D present in cod liver oil.

Although the phosphorus of phytin when supplied in a purified diet is not utilized by the chick, it seems probable that some of it is available when supplied by means of natural products. Such products contain an enzyme called "phytase" which hydrolyzes phytin and sets the phosphorus free. According to the results of McGinnis (1944), however, the presence of phytase in natural products does not increase the availability of the phosphorus of phytin since he obtained no better results with wheat bran, an excellent source of phytase, than with autoclaved wheat bran in which phytase had been destroyed. No explanation, therefore, is available at present for the seemingly better availability of phytin phosphorus when supplied in natural ingredients than when supplied in a purified ration.

The minimum calcium requirement of growing chicks has been reported to be between 0.66 and 0.86 per cent of the ration by Bethke, Kennard, and Kick (1929), at about 0.71 to 0.75 per cent by Hart and associates (1930), and at approximately 0.66 per cent by Wilgus (1931). The minimum calcium requirement of chicks appears, therefore, to be approximately 0.7 per cent.

The minimum phosphorus requirement of growing chicks has been reported by Bethke, Kennard, Kick, and Zinzalian (1929) to be between 0.37 and 0.6 per cent, by Hart and associates (1930) about 0.3 to 0.42 per cent, by Wilgus (1931) 0.5 per cent or less, by Supplee (1935) between 0.26 and 0.5 per cent, and by Watkins and Mitchell (1936) less than 0.5 per cent. More recently Gillis, Norris, and Heuser (unpublished results, Cornell University) found that 0.4 per cent of readily available phosphorus promotes normal bone formation and good growth in young chicks. It appears, therefore, that the minimum phosphorus requirement of chicks is approximately 0.4 per cent, but that to get satisfactory bone development at this level the phosphorus must be present in the ration in a highly available form.

In determining the vitamin D requirements of growing chicks, Murphy, Hunter, and Knandel (1934, 1936) found that the minimum requirement

was between 20 and 40 units per 100 grams of ration. The experimental ration contained 1.47 per cent of calcium and 0.92 per cent of phosphorus. By using experimental rations containing from 2.5 to 2.6 per cent of calcium and 0.9 per cent of phosphorus, Carver and associates (1934) found that the minimum requirement of growing chicks was approximately 20 units of vitamin D per 100 grams. The basal diet used in determining the vitamin D potency of vitamin D preparations to be used for poultry feed contains from 0.9 to 1.0 per cent of calcium, and from 0.85 to 0.9 per cent of phosphorus. In view of the experimental work on the minimum calcium and phosphorus requirements of chicks, the work on the minimum vitamin D requirements, and the method of determining the vitamin D potency of products to be used for feeding poultry, the Poultry Subcommittee of the Committee on Animal Nutrition of the National Research Council set the calcium allowance of chicks at 1.0 per cent and the phosphorus allowance at 0.6 per cent. These recommendations allow approximately a 50 per cent margin of safety over the minimum requirements determined experimentally. The Subcommittee, in addition, specified that the ration must contain at least 0.2 per cent of phosphorus in the inorganic form in order to be certain that not all of the phosphorus in the ration is supplied by means of feedstuffs of vegetable origin. The calcium and phosphorus allowances suggested by the National Research Council are given in Table 2.

Norris and associates (1934), in an extensive investigation of the calcium and phosphorus requirements of laying hens, found that 1.65 per cent of calcium was just sufficient to meet the requirements as judged by egg production, egg shell strength, egg shell ash, and by the levels of calcium and phosphorus in the blood. They concluded that the optimum calcium requirement was about 1.8 per cent. In this work, when 3.33 per cent of calcium was included in the ration a decrease in egg production was obtained, and the hens failed to maintain their weight as well as those fed somewhat lower levels of calcium. Gutowska and Parkhurst (1942) have reported that a ration containing 3.9 per cent of calcium affected egg production detrimentally, but that hatchability and fertility were not adversely affected. The phosphorus content of the ration used in the investigation was 0.75 per cent. Titus and associates (1937), on the other hand, found that levels of calcium varying from 4.05 to 5.4 per cent brought about a decrease in hatchability as well as a decrease in egg production.

The results of Evans and Carver (1941), who studied the calcium and phosphorus requirement of White Leghorn pullets kept in laying cages, show that the best egg production was obtained when the pullets received 2.5 per cent of calcium. It is difficult to reconcile the discrepancy between the results of Norris and associates (1934) and those of Evans and Carver (1941), although it may be due to the short experimental period used by the latter

investigators. Their work was restricted to one experiment covering a period of sixteen weeks, whereas that of Norris and associates included four experiments covering an experimental period of forty weeks each.

Norris and associates (1934), Miller and Bearse (1934), and Evans, Carver, and Brant (1944) have studied the phosphorus requirements of laying hens. Norris and associates, using a ration containing 1.8 per cent of calcium, found that the minimum phosphorus requirement was 0.75 per cent of the diet. Miller and Bearse, using rations varying in calcium from 2.23 to 3.03 per cent, obtained the highest egg production with 0.8 per cent of phosphorus. Evans and associates obtained higher egg production with 0.8 per cent of phosphorus in the ration than when 0.6 per cent was supplied.

An exceptionally high phosphorus turnover is a characteristic of the rapidly laying hen. Not only is the excretion of phosphorus through the usual channels increased, but, in addition, appreciable amounts are deposited in the egg. Phosphorus is found in the egg yolk as a constituent of lecithin and vitellin. Phosphorus is concerned indirectly in egg shell formation although it comprises less than one per cent of the shell. Lorenz, Perlman, and Chaikoff (1943) have followed the deposition of phosphorus in the egg by the use of radioactive phosphorus. Norris and associates (1933) found that egg production per hen was not only decreased when the phosphorus content of the diet was 0.5 per cent, but also that the amount of ash in the egg shell was decreased. Halnan (1925) has pointed out that egg production is associated with increased phosphorus catabolism, and that during egg production the phosphorus lost from the body is much greater than that contained in the eggs laid. Halnan's observations have been confirmed by Common (1932), who found that egg production was correlated with relatively heavy excretion of phosphorus in the feces.

The Poultry Subcommittee of the Committee on Animal Nutrition of the National Research Council has set the calcium requirement of laying hens at 2.25 per cent, thus providing for a margin of safety over the optimum level found by Norris and associates. The phosphorus allowance was set at 0.75 per cent in view of the close agreement between the results of the three groups of investigators. The Subcommittee specified that inorganic phosphorus should constitute 0.2 per cent of the total feed.

The calcium and phosphorus requirements of turkey poults have been studied by Mussehl and Ackerson (1935). They found, in general, that to obtain maximum growth and maximum bone ash in turkey poults, levels of calcium varying from 1.45 to 1.98 per cent and levels of phosphorus varying from 0.63 to 1.02 per cent were required in a ration containing 1.0 per cent of cod liver oil. When no vitamin D or exposure to ultraviolet energy was provided, slightly better growth and bone ash were obtained when the ration contained 2.3 per cent of calcium. Hammond, McClure, and Kellogg

(1944) have reported that 0.6 per cent phosphorus is adequate for growing turkeys and that under favorable conditions as little as 0.5 per cent may be fed without detrimental effect. No studies appear to have been conducted on the calcium and phosphorus requirements of breeding turkeys.

The Poultry Subcommittee of the National Research Council has set the calcium requirements of turkey poults and growing turkeys at 2.0 per cent of the ration and the phosphorus requirement at 1.0 per cent. Because of the more rapid rate of growth of turkey poults, the Subcommittee suggested that the ration contain 0.4 per cent of inorganic phosphorus instead of the lower level suggested for growing chicks. The Subcommittee set the calcium and phosphorus requirements of breeding turkeys at 2.25 and 0.75 per cent of the ration, respectively, in spite of the lack of experimental evidence. This was based upon the fact that breeding chicken rations have promoted satisfactory egg production in breeding turkeys.

*Magnesium.* Magnesium is closely associated with calcium and phosphorus in the body. It is essential for bone formation, about two-thirds of the magnesium in the body being present in the bone, chiefly as a carbonate. It is necessary for carbohydrate metabolism, and activates the enzyme, phosphatase. Egg shells contain about 1.4 per cent of magnesium.

Although magnesium is an essential mineral element, it is present in sufficient amounts in ordinary feedstuffs so that all practical poultry rations contain enough to meet the requirement. It is possible, however, to formulate rations which contain excess magnesium with the result that detrimental effects are produced. Buckner, Martin, and Insko (1932) found that the addition of magnesium carbonate to chick rations in amounts sufficient to raise the magnesium level from 0.76 to 7.05 per cent upset the calcium and phosphorus balance, resulting in deformed bones with low ash and calcium content. A similar effect on bone ash has been reported by Mussehl and co-workers (1930), while Schaible and associates (1933) obtained perosis with magnesium carbonate additions. Alder (1927) found that the use of dolomite limestone containing a high percentage of magnesium for a period of four months caused egg production to decrease and the shells of the eggs to become progressively thinner. Nearly every hen in the dolomite pen developed diarrhea as indicated by the droppings and badly soiled condition of the feathers on the abdomen. The hens also became extremely irritable and easily frightened. All of these conditions cleared up within a short time after substituting a high-grade limestone for the dolomite. Work with several species of animals, however, indicates that magnesium is less harmful when the ration contains liberal amounts of calcium and phosphorus than when these elements are fed at a marginal level.

The magnesium requirement of the chick has been studied by Almquist (1942b), who found it to be approximately 0.04 per cent during the first

few weeks of life. On the basal ration, the chicks grew slowly for approximately one week, then ceased growing and became lethargic. When disturbed, these chicks frequently passed into a brief convulsion accompanied by gasping, and finally into a comatose state which sometimes ended in death.

*Sodium and chlorine (salt)*. Sodium as chloride, carbonate, and phosphate is found chiefly in the blood and body fluids. Sodium chloride is the chief inorganic constituent of the blood plasma, and is presumably the source of chlorine in the hydrochloric acid of the gastric juice. Sodium is connected intimately with the regulation of the hydrogen ion concentration of the blood. Sodium, along with potassium and calcium in proper balance, is essential for heart activity.

The addition of salt to poultry mash mixtures is common practice and probably, in most instances, is necessary to optimum growth and production. Mitchell and Carman (1926) have reported that chicks fed a cereal ration containing no added salt showed retarded growth with decreased efficiency of food utilization. They concluded that the retarded growth was due to a deficiency of sodium rather than of chlorine. These results have been confirmed by Prentice (1933a), who also later found (1933b) that a lack of salt in the ration of laying hens resulted in decreased egg production and egg size, loss of weight, and cannibalism. Sjollema (1935) and Halpin, Holmes, and Hart (1934) obtained better growth in chicks when fed rations containing from 0.5 to 1.0 per cent of salt.

According to the Subcommittee on Poultry Nutrition of the National Research Council, 0.5 per cent of salt should be added to the rations of both chickens and turkeys. This represents added salt and not salt already present in the ingredients of the mash mixture. When the ration is composed of both mash and grain, the mash mixture should contain 1.0 per cent of added salt in order to provide approximately 0.5 per cent in the entire ration.

Excessive amounts of salt in the ration are toxic to chickens. Suffran (1909) studied the toxic effects of salt and found that the lethal dose was approximately 4 grams per kilogram of body weight. Quigley and Waite (1932) fed chicks rations containing from 1.0 to 15.0 per cent of salt and found that levels of 8.0 per cent or greater depressed growth. These workers confirmed the observation of Suffran that the minimum lethal single dose was 4 grams per kilogram of body weight. Mitchell, Card, and Carman (1926) found that chickens from 9 to 21 weeks of age could be fed as much as 8.0 per cent of salt in the ration without any detrimental effects on their rate of growth and physical condition, after the chickens became accustomed to the ration. A daily intake of 6 to 8 grams exerted no harmful effects on these chickens. Torrey and Graham (1935) have reported that ducks are more susceptible to salt poisoning than chickens.

The feeding of excessive amounts of salt causes increased consumption of water and the excretion of watery droppings. The symptoms of salt intoxication are inability to stand, intense thirst, pronounced muscular weakness, and convulsive movements preceding death. Autopsy has revealed lesions in many organs, but particularly hemorrhages and severe congestion in the gastro-intestinal tract, muscles, liver, and lungs.

Halpin (1942) has recommended salt as a means of preventing cannibalism. The treatment consists of dissolving a tablespoon of salt in each gallon of drinking water for one forenoon, and 3 days later repeating the treatment for another half day. Field experience indicates, however, that this method of preventing cannibalism is frequently not successful.

*Potassium.* Potassium is an essential element for both plants and animals. As a result it is widely distributed in feedstuffs of both plant and animal origin, and although the requirement of poultry for this element is rather high, there seems little likelihood of a deficiency occurring in practical poultry rations. In formulating experimental diets of purified ingredients, however, it is necessary to add fairly large amounts of potassium salts.

Potassium, in contrast to sodium, is found primarily in the cells of the body rather than in the body fluids. The soft tissues of the fowl contain more than three times as much potassium as sodium. The sodium and potassium content of the bones are approximately the same. Although the concentration of potassium in the body is high, its fundamental role is not well understood. It is necessary for normal heart activity where it exerts an effect opposite to that of calcium, reducing the contractility of the heart muscle and favoring relaxation. Potassium ions also appear to increase membrane permeability. It is sometimes stated that high intakes of potassium increase the sodium requirements of animals due to increased excretion of the latter element. The evidence on this point is contradictory, however.

Ben Dor (1941) has reported that chicks deficient in potassium exhibit high mortality and retarded growth. He concluded that the chick requires 0.17 per cent potassium in the diet. The potassium requirements of the chick have also been studied by Gillis (unpublished results, Cornell University), who found that 0.16 per cent is sufficient to prevent mortality but that for most rapid growth chicks require from 0.20 to 0.24 per cent potassium.

*Manganese.* Manganese is one of the so-called "trace elements" required by poultry. Although needed in only minute amounts it is one of the mineral elements of major importance in poultry nutrition. Manganese deficiency in growing chicks results in retarded growth, the leg deformity termed perosis, an abnormal shortening of the wing and leg bones, lower ash content of the bones, and reduced phosphatase values in blood and bones. In the laying hen a deficiency may result in failure to maintain weight, lowered

egg production, reduced egg shell strength and egg shell ash, and greatly decreased hatchability with the production of nutritional chondrodystrophy in the embryos. Offspring from hens fed a manganese deficient diet exhibit ataxia and micromelia.

The first abnormality in poultry to be associated with manganese deficiency was perosis. Perosis, or slipped tendon as it is frequently called, is an anatomical deformity of the leg bones of young chickens, turkeys, pheasants, grouse, and quail. The symptoms generally found are gross enlargement of the tibial-metatarsal joint, twisting or bending of the distal end of the tibia and of the proximal end of the metatarsus, and finally slipping of the gastrocnemius tendon from its condyles (Fig. 6.3). The latter symptom causes complete crippling in the affected leg, and if both legs are so affected death usually results due to the inability of the bird to secure food and water.

Early observers of perosis noted that it was most likely to occur under the crowded conditions of confinement rearing, that rations having a high mineral content tended to aggravate the condition, and that heavy breeds were more susceptible than light breds. Wilgus, Norris, and Heuser (1936, 1937a) first reported that the deformity is associated with a manganese deficiency. These workers found that perosis in chicks was almost entirely prevented by adding 25 p.p.m. manganese to a diet already containing 10 p.p.m. They also showed that the perosis-preventing property of certain feedstuffs is correlated with their manganese content. These findings were almost immediately confirmed by a number of other investigators.



FIG. 6.3. An extreme case of perosis. The most common causative factor of this disease is manganese deficiency.

The amount of manganese required by chicks varies with different breeds and strains. Wilgus and associates (1937a) reported 35 p.p.m. to be adequate for the crossbred chicks used in their experiments. Insko, Lyons, and Martin (1938) found 36 or 37 p.p.m. to be adequate for growth and the prevention of perosis in Rhode Island Red chicks. Gallup and Norris (1939a) reported that approximately 50 p.p.m. were required by New Hampshire chicks, while 30 p.p.m. completely prevented the disorder in

White Leghorn chicks. Under practical conditions, it is usually recommended that rations for both chicks and poults contain 50 p.p.m. manganese.

When manganese is injected into the growing chicken, less is required to prevent perosis than when it is fed orally (Caskey and Norris, 1939). This is particularly true when the diet contains excess calcium and phosphorus. The greater effectiveness of injected manganese is due to the inefficient absorption of this mineral from the digestive tract. Manganese is even less readily absorbed when an excess of other minerals is present in the diet, particularly calcium, phosphorus, and iron, because they tend to form insoluble combinations with the manganese or precipitates which adsorb it. The tendency of excess phosphorus and other minerals to aggravate the occurrence of perosis was recognized for a considerable time before manganese was associated with the disorder (Insko, Sowell, and Lyons, 1934; Hammond, 1935; Heller and Penquite, 1936).

Besides manganese, there are several organic factors concerned in the prevention of perosis. Biotin, choline, and folic acid have definitely been shown to be concerned, and there is strong evidence that riboflavin and niacin are likewise implicated particularly in the case of turkey poults.

In addition to its perosis-preventing properties, manganese is necessary for the formation of normal bones. Wilgus, Norris, and Heuser (1937b) reported that frequently the leg bones of chicks fed perosis-producing diets were thickened and shortened. Gallup and Norris (1938) reported that the leg bones of chicks on a manganese deficient diet were 7 or 8 per cent shorter than those of chicks of the same age, sex, and weight receiving adequate manganese. Despite the fact that the bones of the deficient chicks were perceptibly thicker and shorter than the controls, calcification appeared to be normal. In a more extensive study, Caskey, Gallup, and Norris (1939) reported that manganese deficiency in the diet of chicks resulted in a highly significant shortening of the bones of the legs and wings as well as a shortening of the spinal column. They also found that the ash content of the bones of the deficient chicks was significantly lower than that of chicks fed an adequate diet.

The evidence that manganese plays an important role in bone formation is supported by the observations of Wiese and associates (1939, 1941) that the bone and blood phosphatase, as well as ester phosphorus of the blood, are depressed in manganese deficiency in the chick. The activity of the enzyme phosphatase is vitally concerned in the metabolism of phosphorus and thereby in the formation of bone. Combs and associates (1942) have confirmed the observation that manganese deficiency lowers bone phosphatase activity. They have also shown that the abnormally high phosphatase activity associated with rickets is reduced by a concurrent manganese deficiency. These results, together with those of Caskey, Gallup, and Norris

(1939), suggest that in manganese deficiency the lowering of the phosphatase level retards bone development. This results in shorter bones having a lower ash content and a weakened union between the epiphysis and the diaphysis. It is probable that manganese deficiency in the diet causes a disproportionately greater retardation in the development of bones in chicks during growth than of other tissues of the body. As a result of this the bones are not strong enough to support the weight of the body, and malformation occurs.

Dietary manganese is as important for mature fowls as it is for the young



FIG. 6.4. The shortened limbs and parrot beak are typical of the effect of manganese deficiency upon embryos.

growing stock. Laying and breeding hens are quickly affected by a deficiency of this mineral. Gallup and Norris (1937, 1939b) reported that pullets on a diet containing 13 p.p.m. manganese produced less than half the number of eggs produced by similar pullets on a diet containing 200 p.p.m. They also found that the manganese content of the eggs from the low manganese group was reduced. Fertility was slightly decreased, and the hatchability of eggs was markedly lowered by manganese deficiency. Embryos in the eggs of low manganese content usually died during the final stages of incubation.

Lyons and Insko (1937) also found that manganese deficiency resulted in very low hatchability of fertile eggs and, in addition, observed the production of chondrodystrophy in the embryos (Fig. 6.4). The peak of mortality for such embryos occurred on the twentieth and twenty-first days of incubation. The chondrodystrophic embryos were characterized by very short, thickened legs, short wings, "parrot beak," globular contour of head, protruding abdomen, and retarded down and body growth. Very marked edema was noted in about 75 per cent of these embryos. It was found that the man-

ganese content of the eggs producing chondrodystrophic embryos was much smaller than that of normal eggs. Chondrodystrophy was completely prevented by the injection of 0.03 mg. of manganese into the albumen of deficient eggs or by increasing the manganese in the diet.

Chicks hatched from eggs produced on a diet deficient in manganese sometimes exhibit ataxia (Caskey, Norris, and Heuser, 1944). This is a nervous disorder involving spasms in which the head may be drawn forward and bent underneath the body or retracted over the back. These spasms are most apparent when the chicks are excited. The ataxic chicks may grow normally and reach maturity. However, they retain the short bones characteristic of embryos and newly hatched chicks when the maternal diet is deficient in manganese. This condition of micromelia has been described by Caskey and Norris (1940).

One of the important functions of manganese is the maintenance of the breaking strength of the egg shell. Caskey and Norris (1938) found that the breaking strength of the eggs from lots of hens receiving different levels of manganese increased as the quantity of manganese in the diet increased. The breaking strength averaged 6.6 pounds for a lot receiving 6.5 p.p.m. manganese and 9.3 pounds for a lot receiving 100 p.p.m. The ash content of the egg shell also increases with increase in the manganese content of the diet. Lyons (1939) has described the effect of a low manganese diet on the characteristics of egg shells. Eggs produced on a deficient diet show large areas of poor calcification as indicated by differences in smoothness and translucency. A lowered breaking strength and a smaller percentage of shell were also found.

The manganese requirement of hens is less than that of chicks. Caskey and Norris (1938) found 20 p.p.m. to be sufficient for egg production and hatchability. The results of Schaible, Bandemer, and Davidson (1938) indicated that additional manganese above 39 p.p.m. did not benefit fertility, hatchability, or egg production. Breed and strain differences in requirements have been pointed out by Golding, Schaible, and Davidson (1940), who found that heavy breeds require more manganese than light breeds. The work with hens, therefore, supports the results obtained with chicks, which shows that heavy breeds are more susceptible to manganese deficiency than light breeds. Golding and associates found that 9 p.p.m. of dietary manganese were adequate for hatchability and the prevention of chondrodystrophy but not for egg production in White Leghorn pullets. This amount of manganese was not sufficient for hatchability in Barred Plymouth Rocks, however.

Recent evidence by Couch, James, and Sherwood (1947) indicates that the manganese requirements of hens in their second year of production is greater than that of pullets. They report that New Hampshire pullets re-

quire not more than 41 p.p.m. for egg production and between 41 and 71 p.p.m. for egg shell quality and for fertility and hatchability. On the other hand, New Hampshire hens were found to require approximately 71 p.p.m. of manganese for egg production. The requirements for hatchability and fertility in hens were similar to those of pullets. These workers found that high levels of manganese in the diet lower the vitamin D requirement.

The economic importance of supplying adequate dietary manganese and the low cost of adding this mineral to feeds has resulted in the widespread addition of manganese salts to poultry rations. Several compounds are equally satisfactory for this purpose (Schaible, Bandemer, and Davidson, 1938). Manganous sulfate, chloride, carbonate, potassium permanganate, and manganese dioxide appear to be equally prophylactic. Oxide ores of manganese such as manganite and pyrolusite are comparable to the foregoing compounds; however, the carbonate ore, rhodochrosite, and the silicate ore, rhodonite, are not satisfactory sources of manganese. A common practice among feed manufacturers is to add 4 ounces of manganous sulfate to a ton of mixed feed. This supplies about 25 to 35 p.p.m. of manganese depending on the degree of hydration of the manganous sulfate. Although high concentrations of manganese are poisonous, the amounts encountered in practice are much below the toxic level. More than thirty to forty times the amount required have been fed without apparent harm.

*Iodine.* Traces of iodine are required for normal functioning of the thyroid gland in poultry as well as all other animal species. Most of the iodine in the body is held by the thyroid which has a remarkable affinity for this element. Thyroxin, the hormone secreted by the thyroid, contains approximately 65 per cent iodine and acts as an important regulating agent in body metabolism. When the intake of iodine is suboptimal, the thyroid tissue enlarges and the condition known as goiter results.

The problem of iodine deficiency is confined to areas in which the soil, and consequently the water and feed crops, contain insufficient amounts of this mineral. In the United States these areas are primarily in the Northwest and in the Great Lakes region, but many other sections are either marginal or actually deficient in this respect. To some extent iodine deficiency in poultry has probably been offset by the widespread use of fish meal, oyster shells, and fish oils in poultry rations. These marine products all contain significant amounts of iodine. Nevertheless, goiter has been described in poultry by a number of investigators. In 1928, Welch reported that goiter occurred very commonly in poultry in Montana. He minimized the effect of this condition on the general health and productivity of the affected birds, however. Kernkamp (1925) has given a detailed report of simple colloid goiter in poultry. The problem of goiter in chickens and the associated iodine requirements have been investigated by the Colorado workers

(Patton, Wilgus, and Harshfield, 1939; Wilgus, Harshfield, Patton, Ferris, and Gassner, 1941b; Gassner and Wilgus, 1940). These workers have reported that iodine deficiency results in enlarged thyroids and, in some cases, lower body weight in growing chicks. They estimated that the growing chicken requires 1 p.p.m. of iodine in the diet for optimum growth and the prevention of goiter. They observed congenital goiter in baby chicks hatched from hens receiving 0.025 p.p.m. of iodine in the ration. Goiter in domestic pigeons has been investigated by Hollander and Riddle (1946).

Wilgus and associates (1941a) have observed that soybeans in the diet of poultry have a goitrogenic effect. They found that this effect is partially inactivated by heat, however, and that iodine is a counteractant. Soybean oil meal was also found to be goitrogenic, but no detrimental effects other than that on the thyroid gland were noted in its use in feeding growing chickens.

The iodine requirements of poultry cannot be stated with any degree of certainty. However, it is known that certain levels prevent the appearance of any deficiency symptoms. The addition of iodine supplements to poultry rations in nongoitrogenic areas is contraindicated by a large amount of experimental data. According to the National Research Council (Griem, Hart, Kalkus, and Welch, 1942), the addition of 0.5 per cent of iodized salt to poultry rations is adequate to meet the requirements under all conditions. The Poultry Subcommittee of the National Research Council has set the iodine "allowance" of poultry rations at 0.5 mg. per pound of feed.

It is possible to greatly increase the iodine content of eggs by feeding supplementary iodine to laying hens (Wilder, Bethke, and Record, 1933; Asmundson, Almquist, and Klose, 1936). Inasmuch as iodine is a necessary constituent of the human diet, these "iodized eggs" have sometimes been recommended as a means of supplementing the human dietary. This is impractical, however, since the human requirement can be met much more economically by the use of iodized salt and other foods naturally rich in iodine.

*Iron and copper.* Both iron and copper are necessary for hemoglobin formation. Iron is present in hematin, the iron porphyrin nucleus of hemoglobin. This nucleus is also one of the components of cytochrome and the enzymes, peroxidase and catalase. Copper, on the other hand, while essential for hemoglobin formation, does not enter into its composition. In the absence of copper, dietary iron is absorbed and deposited in the liver and elsewhere. Hemoglobin formation, however, does not occur, and anemia results. Copper is found in the blood cells as hemocuprin. Elvehjem, Hart, and Kemmerer (1929) have shown that both iron and copper are required for hemoglobin synthesis in the chick, and Hart and associates (1929) concluded that practical chick starter rations contain enough iron and copper for the prevention of anemia.

The demand for iron and copper for egg formation is large, as the average egg contains about 1.1 mg. of iron and 0.067 mg. copper. The results of several investigators indicate that the feeding of extra iron and copper to the hen does not increase the iron and copper content of the eggs, although Erikson and associates (1933) found that hens with access to sunshine and bluegrass range produced eggs with higher iron and copper content than hens confined indoors.

The evidence, in general, seems to indicate that practical chick rations contain sufficient iron and copper to prevent the development of anemia. Several investigators, however, have observed that the hemoglobin level falls with the beginning of egg production. This does not appear to be related to the iron and copper content of the diet. In view of the fact that the hemoglobin level rises rapidly with the onset of broodiness, it appears more probable that the low hemoglobin levels which prevail in egg production are due to changes in the hormone mechanisms of the body rather than to iron and copper deficiencies.

It is necessary to avoid the feeding of mineral supplements which contain large excesses of iron due to the tendency of iron to render several other essential elements unavailable.

*Sulfur.* Sulfur is probably the only mineral element required by poultry which must be supplied entirely in the organic form. Actually from the viewpoint of nutrition the intake of sulfur is not a problem, provided the protein is adequate. The amino acids, methionine and cystine, which contain sulfur in organic combination, account for practically all the sulfur utilized by poultry. Traces of sulfur are also supplied by the intake of the vitamins, thiamin and biotin. Elemental sulfur and inorganic sulfates do not appear to have any value nutritionally. According to Hendricks (1933), the feeding of these compounds to laying hens has no apparent effect on the length of the molting period, or upon the growth or character of the new feathers produced.

The amino acids, methionine and cystine, are not the only compounds in the body containing sulfur. All the other sulfur-containing compounds, however, appear to be derived from the catabolism of these amino acids, and many of them appear in the urine as excretory products. Marlowe and King (1936) studied the sulfur content of egg yolk and egg white and found that all of the sulfur in these products was organically bound, and also that nearly all could be accounted for by the cystine and methionine sulfur.

A number of investigators have reported that sulfur is of value in preventing cecal coccidiosis in chicks. Detrimental effects are obtained, however, when large amounts of sulfur are supplied for a prolonged period of time. Holmes, Deobald, and Herrick (1937) have reported that feeding flowers of sulfur to chicks for six weeks, even with cod liver oil, resulted in

the development of rickets. On the other hand, they found that when chicks were exposed to sunlight, 5 per cent of flowers of sulfur failed to promote rickets. Rickets developed when the ration contained 0.5 or 1.0 per cent cod liver oil of a potency of 175–225 U.S.P. units of vitamin D per gram. When the level of cod liver oil was increased to 2.0 per cent, bone ash was nearly normal, although growth was not materially improved.

*Zinc.* Traces of zinc appear,to be necessary for life in all animals. It is a constituent of the enzyme, carbonic anhydrase, and possibly other enzymes and hormones require this mineral for their functioning. Zinc is widely distributed in the feedstuffs consumed by poultry, and there appears to be no likelihood of its becoming a problem in practical poultry nutrition.

*Fluorine.* When appreciable quantities of fluorine are ingested by poultry, harmful effects result. The fluorine tolerance of chickens, however, seems to be definitely higher than that of other species of farm animals. In determining the value of experimental work, it is well to bear in mind that the toxic effects of fluorine are cumulative and that short term experiments with fluorine are seldom reliable.

Halpin and Lamb (1932) have shown that the toxic level of fluorine in the ration of chickens is between 0.035 and 0.070 per cent. Excess fluorine resulted in retarded growth in young chicks, and lower egg production and loss of weight in laying hens. The results of Hauck and associates (1933) indicate that the toxic level of fluorine, as measured by the growth of chicks, lies between 0.068 and 0.136 per cent. Kick, Bethke, and Record (1933) have indicated that the toxic level is between 0.036 and 0.072 per cent. The whole problem of fluorine in animal nutrition has been reviewed by Mitchell (1942).

The Association of American Feed Control Officials has recommended that the fluorine content of minerals or mineral mixtures used in poultry feedstuffs contain not more than 0.60 per cent of fluorine, and that the total fluorine content of poultry rations should not exceed 0.035 per cent. The Committee on Animal Nutrition of the National Research Council has suggested that the permissible level of fluorine be set at 0.015 per cent of the total dry ration for poultry.

The fluorine hazard in poultry feeding, outside of certain local areas, is related largely to the use of fluorine-containing mineral supplements. These minerals are the different varieties of phosphate rock, the superphosphates produced from them, and the phosphatic limestones. Raw rock phosphates generally contain from 3.25 to 4.0 per cent of fluorine, while superphosphate and phosphatic limestone usually contain appreciably less. Heat treatment of the raw rock phosphate or superphosphate is employed to remove the fluorine from these products. Commercially defluorinated phosphate rock and superphosphate as now marketed generally come well within the limits

of permissible fluorine content. These materials provide valuable sources of calcium and phosphorus, but it is well for the prospective buyer to make sure that the fluorine content has been reduced to the recommended limits for mineral supplements. It is possible to obtain defluorinated phosphate products containing as little as 0.02 to 0.03 per cent of fluorine.

*Selenium.* Selenium is present in a number of plants, growing upon certain soils, in concentrations ranging from mere traces to 0.2 per cent. The feeding of these plants or by-products made from them may result in alkali disease or selenium poisoning. Any plant product containing 5 p.p.m. of selenium is potentially dangerous.

Tully and Franke (1935) have reported that chicks supplied a ration containing 65 per cent of grains grown on seleniferous soil showed retarded growth, ruffled feathers, irritability, and delayed and reduced egg production. In later work (Franke and Tully, 1935, 1936) these investigators reported that the eggs produced by hens fed a ration of this character possessed poor hatchability and that almost three-quarters of the embryos in the eggs which failed to hatch were deformed. Franke, Moxon, Poley, and Tully (1936) showed that these effects were produced by selenium when it was injected into the eggs before incubation. Poley and Moxon (1938) fed laying rations containing 2.5, 5.0, and 10.0 p.p.m. of selenium. At the intermediate level hatchability was not appreciably affected, but some chicks had wiry down. The high level, on the other hand, reduced hatchability to zero, the dead embryos showing short upper beaks, edema of head and neck, missing toes and eyes, wiry down and other characteristics of selenium poisoning. No effects on body weight, egg production, or fertility were observed. Moxon and Poley (1938) investigated the selenium content of the body of these hens. This was increased proportionately to the amount in the ration. The selenium content of the eggs was also increased in proportion to the amount supplied in the ration.

## WATER

The simple chemical compound, water, is of unequaled importance in the metabolism of all animals. Water holds this unique position in nutrition mainly because of its physical properties. Due to its solvent and polar properties, it acts as a transport medium for all other nutrients and products of metabolism and enhances cell reactions. Because of its high specific heat it can absorb the heat of reaction produced in the burning of carbohydrates and fats with little rise in temperature. Water evaporates readily, removing many calories of heat from the body as latent heat of vaporization. These and the many other functions of water make it evident why the animal body is able to exist much longer without food than it can without water.

*Importance of a continuous water supply for poultry.* It is not possible

to supply sufficient water to poultry by placing it before the birds only once or twice a day, as is done with larger farm animals. Since chickens drink only a small amount of water at one time, they must have access to a continuous water supply. During severe weather it is also necessary to make some provision which will keep the water from freezing.

A deficient amount of water for poultry results in decreased growth and egg production. Hammond (1944) has observed that a lack of water causes the development of loose, slimy gizzard linings which accompany early, nonspecific mortality in turkey poults. He also observed that, unlike the day-old chick which begins to eat and drink without being taught, many day-old poults fail to learn to eat or drink even when they are in presence of other poults that are eating and drinking.

## REFERENCES

Ackerson, C. W., Blish, M. J., and Mussehl, F. E.: 1939. The utilization of food elements by growing chicks. 6. The influence of the protein level of the ration on the growth of chicks. Nebr. Agr. Exper. Sta., Res. Bul. 108.

Alder, B.: 1927. The use of calcite and other natural deposits of calcium carbonate in the ration of laying hens. Proc. Third World's Poultry Cong. P. 231.

Almquist, H. J.: 1942a. The amino acid requirements and protein metabolism of the avian organism. Fed. Proc. 1:269.

——: 1942b. Magnesium requirement of the chick. Proc. Soc. Exper. Biol. and Med. 49:544.

——: 1945a. Effective use of proteins in the nutrition of the chick. Trans. Am. Assn. Cer. Chem. 3:158.

——: 1945b. Proteins and amino acids in animal nutrition. Bul. F. E. Booth Company, Inc., San Francisco.

—— and Asmundson, V. S.: 1944. High protein mashes for broilers. Poultry Sci. 23:67.

—— and Grau, C. R.: 1944. The amino acid requirements of the chick. Jour. Nutr. 28:325.

Anonymous: 1935. Chicken raising experiments. Queensland Agr. Jour. 44:425.

Asmundson, V. S., Almquist, H. J., and Klose, A. A.: 1936. Effect of different forms of iodine on laying hens. Jour. Nutr. 12:1.

Ben Dor, Ben-Ami: 1941. Requirements of potassium by the chick. Proc. Soc. Exper. Biol. and Med. 46:341.

Bethke, R. M., Kennard, D. C., and Kick,·C. H.: 1929. The availability of calcium in calcium salts and minerals for bone formation in the growing chick. Poultry Sci. 9:45.

——, Kennard, D. C., Kick, C. H., and Zinzalian, G.: 1929. The calcium-phosphorus relationship in the nutrition of the growing chick. Poultry Sci. 8:257.

Bird, H. R., Rubin, M., Whitson, D., and Haynes, S. K.: 1946. Effectiveness of dietary supplements in increasing hatchability of eggs and viability of progeny of hens fed a diet containing a high level of soybean oil meal. Poultry Sci. 25:285.

Block, R. J., and Bolling, D.: 1945. The Amino Acid Composition of Proteins and Foods. Charles C. Thomas, Springfield, Ill.

——, Jones, D. B., and Gersdorff, C. E. F.: 1934. The effect of dry heat and dilute alkali on the lysine content of casein. Jour. Biol. Chem. 105:667.

Boatner, C. H., and Hall, C. M.: 1946. Pigment glands of cottonseed. I. Behavior of the glands toward organic solvents. Oil and Soap 23:123.

Bronkhorst, J. J.: 1938. The influence of the protein level of diet on the growth, egg production, egg weight and mortality of Single Comb White Leghorn pullets. Onderstepoort Vet. Sci. 10:469.

Buckner, G. D., Martin, J. H., and Insko, Jr., W. M.: 1932. The effect of magnesium carbonate when added to diets of growing chicks. Poultry Sci. 11:58.

Burr, G. O., and Burr, M. M.: 1930. On the nature and role of the fatty acids essential in nutrition. Jour. Biol. Chem. 86:587.

Byerly, T. C., Titus, H. W., and Ellis, N. R.: 1933. Production and hatchability of eggs as affected by different kinds and quantities of proteins in the diet of laying hens. Jour. Agr. Res. 46:1.

Carver, J. S., Heiman, V., Cook, J. W., and St. John, J. L.: 1939. The protein requirements of White Leghorn pullets. Wash. Agr. Exper. Sta., Bul. 383.

——, Robertson, E. I., Brazie, D., Johnson, R. H., and St. John, J. L.: 1934. The vitamin D requirements of chickens. Wash. Agr. Exper. Sta., Bul. 299.

————, St. John, J. L., Aspinall, T. E., and Flor, I. H.: 1932. Protein requirements of chickens. Poultry Sci. 11:45.

Caskey, C. D., Gallup, W. D., and Norris, L. C.: 1939. The need for manganese in the bone development of the chick. Jour. Nutr. 17:407.

———— and Norris, L. C.: 1938. Further studies on the role of manganese in poultry nutrition. Poultry Sci. 17:433.

———— and Norris, L. C.: 1939. Relative effectiveness of ingested and injected manganese in preventing perosis. Proc. Soc. Exper. Biol. and Med. 40:590.

———— and Norris, L. C.: 1940. Micromelia in adult fowl caused by manganese deficiency during embryonic development. Proc. Soc. Exper. Biol. and Med. 44:332.

————, Norris, L. C., and Heuser, G. F.: 1944. A chronic congenital ataxia in chicks due to manganese deficiency in the maternal diet. Poultry Sci. 23:516.

Combs, G. F., Norris, L. C., and Heuser, G. F.: 1942. The interrelationship of manganese, phosphatase, and vitamin D in bone development. Jour. Nutr. 23:131.

Common, R. H.: 1932. Mineral balance studies on poultry. Jour. Agr. Sci. 22:576.

Couch, J. R., James, L. E., and Sherwood, R. M.: 1947. The effect of different levels of manganese and different amounts of vitamin D in the diet of hens and of pullets. Poultry Sci. 26:30.

Drea, W. F.: 1935. Spectrum analysis of hen eggs and chick tissues. Jour. Nutr. 10:351.

Elvehjem, C. A., Hart, E. B., and Kemmerer, A. R.: 1929. The relation of iron and copper to hemoglobin synthesis in the chick. Jour. Biol. Chem. 84:131.

Erikson, S. E., Boyden, R. E., Martin, J. H., and Insko, Jr., W. M.: 1933. The iron and copper content of egg yolk. Ky. Agr. Exper. Sta., Bul. 342:135.

Evans, R. J., and Carver, J. S.: 1941. The effect of mineral metabolism on egg shell quality. Wash. Agr. Exper. Sta., Bul. 410:92.

————, Carver, J. S., and Brant, A. W.: 1944. The influence of dietary factors on egg shell quality. I. Phosphorus. Poultry Sci. 23:9.

Franke, K. W., and Tully, W. C.: 1935. A new toxicant occurring naturally in certain samples of plant foodstuffs. V. Low hatchability due to deformities in chicks. Poultry Sci. 14:273.

———— and Tully, W. C.: 1936. A new toxicant occurring naturally in certain samples of plant foodstuffs. VII. Low hatchability due to deformities in chicks produced from eggs obtained from chickens of known history. Poultry Sci. 15:316.

————, Moxon, A. L., Poley, W. E., and Tully, W. C.: 1936. Monstrosities produced by the injection of selenium salts into hens' eggs. Anat. Record 65:15.

Fraps, G. S.: 1946. Composition and productive energy of poultry feeds and rations. Tex. Agr. Exper. Sta., Bul. 678.

Fritz, J. C., Halpin, J. L., and Hooper, J. H.: 1947. Studies on the nutritional requirements of poults. Poultry Sci. 26:78.

————, Hooper, J. H., Halpin, J. L., and Moore, H. P.: 1946. Failure of feather pigmentation in bronze poults due to lysine deficiency. Jour. Nutr. 31:387.

Gallup, W. D., and Norris, L. C.: 1937. Studies on the importance of manganese in the nutrition of poultry. Poultry Sci. 16:351.

———— and Norris, L. C.: 1938. The essentialness of manganese for the normal development of bone. Science 87:18.

–———— and Norris, L. C.: 1939a. The amount of manganese required to prevent perosis in the chick. Poultry Sci. 18:76.

———— and Norris, L. C.: 1939b. The effect of a deficiency of manganese in the diet of the hen. Poultry Sci. 18:83.

Gassner, F. X., and Wilgus, Jr., H. S.: 1940. Congenital goiter in chicks. Poultry Sci. 19:349.

Gericke, A. M., and Platt, C. S.: 1932. Feather development in Barred Plymouth Rock chicks. N. J. Agr. Exper. Sta., Bul. 543.

Golding, W. V., Schaible, P. J., and Davidson, J. A.: 1940. A breed difference in the manganese requirement of laying hens. Poultry Sci. 19:263.

Grau, C. R., Kratzer, F. H., and Asmundson, V. S.: 1946. The lysine requirements of poults and chicks. Poultry Sci. 25:529.

Greaves, E. O., and Morgan, A. F.: 1934. Nutritive value of raw and heated casein with and without added amino acids. Proc. Soc. Exper. Biol. and Med. 31:506.

Griem, W. B., Hart, E. B., Kalkus, J. W., and Welch, H.: 1942. Iodine—its necessity and stabilization. Nat. Res. Council Reprint and Circ. Series. No. 111.

Groschke, A. C., Rubin, M., and Bird, H. R.: 1947. Gland-free cottonseed meal as a protein supplement for chickens. Poultry Sci. 26:310.

Gutowska, M. S., and Parkhurst, R. T.: 1942. Studies in mineral nutrition of laying hens. II. Excess of calcium in the diet. Poultry Sci. 21:321.

Halnan, E. T.: 1925. The calcium, phosphorus and nitrogen balance of the nonlaying and laying pullet. Jour. Nat. Poultry Inst. 10:410.

————: 1936. The role of minerals in poultry nutrition. Proc. Sixth World's Poultry Cong. I:53.

Halpin, J. G.: 1942. Besides salt, what's needed to prevent cannibalism? Wis. Agr. Exper. Sta., Bul. 455:15.

Halpin, J. G., Holmes, C. E., and Hart, E. B.: 1934. Salt requirements of growing chicks. Poultry
    Sci 13:308.
———— and Lamb, A. R.: 1932. The effect of ground phosphate rock fed at various levels on the
    growth of chicks and on egg production. Poultry Sci. 11:5.
Ham, W. E., Sandstedt, R. M., and Mussehl, F. E.: 1945. The proteolytic inhibiting substance
    in the extract from unheated soybean meal and its effect upon growth in chicks. Jour. Biol.
    Chem. 161:635.
Hammond, J. C.: 1935. Inorganic phosphorus and perosis. Poultry Sci. 14:302.
————: 1944. Lack of water a cause of loose, slimy gizzard linings accompanying early mortality
    in poults. Poultry Sci. 23:477.
————, McClure, H. E., and Kellogg, W. L.: 1944. The minimum phosphorus requirements of
    growing turkeys. Poultry Sci. 23:239.
Hart, E. B., Elvehjem, C. A., Kemmerer, A. R., and Halpin, J. G.: 1929. Does the practical chick
    ration need iron and copper additions to insure normal hemoglobin building? Poultry
    Sci. 9:92.
————, Scott, H. T., Kline, O. L., and Halpin, J. G.: 1930. The calcium-phosphorus ratio in the
    nutrition of the growing chick. Poultry Sci. 9:296.
Hartwick, H.: 1940. Ueber das Vorkommen der Visceralgicht (Eingeweidegicht) bei Junghühnern
    und Hühnerkücken. Tierärztl. Rundschau 46:812.
Hauck, H. M., Steenbock, H., Lowe, J. T., and Halpin, J. G.: 1933. Effect of fluorine on growth,
    calcification and parathyroids in the chicken. Poultry Sci. 12:242.
Heiman, V., Carver, J. S., and St. John, J. L.: 1936. The protein requirement of laying hens.
    Wash. Agr. Exper. Sta., Bul. 331.
Heller, V. G., and Penquite, R.: 1936. Factors producing and preventing perosis. Poultry Sci.
    15:424.
Hendricks, W. A.: 1933. A biometric study of molt in White Leghorn hens. Poultry Sci. 12:287.
Henneberg, W., and Stohmann, F.: 1860, 1865. Beiträge zur Begründung einer rationellen
    Fütterung der Wiederkäuer. I, II, Schwetschke u. Sohn, Brunswick.
Heuser, G. F.: 1936. The protein requirements of laying hens. Proc. Sixth World's Poultry
    Cong. I:276.
————: 1941. Protein in poultry nutrition—a review. Poultry Sci. 20:362.
————: 1946. Feeding Poultry. John Wiley and Sons, Inc., New York. Pp. 451–57.
———— and Norris, L. C.: 1933. The influence of the protein level on the growth of chickens
    and its relation to subsequent behavior. Proc. Fifth World's Poultry Cong. II:551.
————, Norris, L. C., and McGinnis, J.: 1946. Vegetable protein concentrates fed alone and in
    combination with soybean oil meal and fish meal as the chief supplementary protein in chick
    starting rations. Poultry Sci. 25:130.
Hibbert, H.: 1942. Lignin. Ann. Rev. Biochem. 11:183.
Hill, D. C.: 1944. Protein in poultry nutrition. Scient. Agr. 24:551.
Hollander, W. F., and Riddle, O.: 1946. Goiter in domestic pigeons. Poultry Sci. 25:20.
Holmes, C. E., Deobald, H. J., and Herrick, C. A.: 1937. Sulphur and rickets. Poultry Sci. 16:366.
Hull, T. A., and Rettger, L.: 1917. Influence of milk and carbohydrate feeding on the character of
    intestinal flora. Jour. Bact. 2:47.
Insko, Jr., W. M., Lyons, M., and Martin, J. H.: 1938. The quantitative requirement of the
    growing chick for manganese. Jour. Nutr. 15:621.
————, Sowell, D. F., and Lyons, M.: 1934. Is phosphorus a causative factor in the production
    of slipped tendon? Poultry Sci. 13:370.
Kernkamp, H. C. H.: 1925. Goiter in poultry. Jour. Am. Vet. Med. Assn. 67:223.
Kick, C. H., Bethke, R. M., and Record, P. R.: 1933. Effect of fluorine in the nutrition of the
    chick. Poultry Sci. 12:382.
Lorenz, F. W., Perlman, I., and Chaikoff, I. L.: 1943. Phosphorus deposition in the egg as
    measured with radioactive phosphorus. Am. Jour. Physiol. 138:318.
Lyons, M.: 1939. Some effects of manganese on eggshell quality. Ark. Agr. Exper. Sta., Bul. 374.
———— and Insko, Jr., W. M.: 1937. Chondrodystrophy in the chick embryo produced by
    manganese deficiency in the diet of the hen. Ky. Agr. Exper. Sta., Bul. 371.
Margolf, P. H.: 1929. The effects of various protein-carbohydrate ratios upon the mortality,
    growth and condition of Single Comb White Leghorn chicks. 42nd Ann. Rep. Pa. St. Coll.,
    Pa. Agr. Exper. Sta., Bul. 243:28.
Marlow, H. W., and King, H. H.: 1936. Sulfur in eggs. Poultry Sci. 15:377.
Mattill, H. A.: 1927. The oxidative destruction of vitamins A and E and the protective action
    of certain vegetable oils. Jour. Am. Med. Assn. 89:1505.
Mayall, G.: 1929. Visceral gout in poultry. Vet. Jour. 85:230.
Maynard, L. A.: 1947. Animal Nutrition. Second Ed. McGraw-Hill Book Company, New York.
    P. 48.
McConachie, J. D., Graham, Jr., W. R., and Branion, H. D.: 1935. A study of the protein
    requirements of growing chicks. Scient. Agr. 15:754.
McGinnis, J.: 1944. Studies on the utilization by the chick of phosphorus supplied entirely from
    plant sources. Thesis, Cornell Univ.

——— and Menzies, V. H.: 1946. Effect of in vitro enzymatic digestion of raw soybean flakes on chick growth. Poultry Sci. 25:538.

———, Norris, L. C., and Heuser, G. F.: 1944a. Influence of diet on chick growth-promoting and antiperotic properties of betaine, methionine and choline. Proc. Soc. Exper. Biol. and Med. 56:197.

———, Norris, L. C., and Heuser, G. F.: 1944b. Poor utilization of phosphorus in cereals and legumes by chicks for bone development. Poultry Sci. 23:157.

Miller, M. W., and Bearse, G. E.: 1934. Phosphorus requirements of laying hens. Wash. Agr. Exper. Sta., Bul. 306.

Milne, H. I.: 1932. Protein requirements of growing chicks. Scient. Agr. 12:604.

Mitchell, H. H.: 1942. The fluorine problem in livestock feeding. Nat. Res. Council Reprint and Circ. Series. No. 113.

———, Card, L. E., and Carman, G. G.: 1926. The toxicity of salt for chickens. Ill. Agr. Exper. Sta., Bul. 279.

———, Card, L. E., and Hamilton, T. S.: 1926. The growth of White Plymouth Rock chickens. Ill. Agr. Exper. Sta., Bul. 278.

———, Card, L. E., and Hamilton, T. S.: 1931. A technical study of the growth of White Leghorn chickens. Ill. Agr. Exper. Sta., Bul. 367:83.

——— and Carman, G. G.: 1926. Does the addition of sodium chloride increase the value of a corn ration for growing animals? Jour. Biol. Chem. 68:165.

Morris, L., Thompson, R. B., and Heller, V. G.: 1932. The effect of varying the amounts of protein in the poultry ration on chick growth and subsequent egg production. Poultry Sci. 11:364.

Moxon, A. L., and Poley, W. E.: 1938. The relation of selenium content of grains in the ration to the selenium content of poultry carcass and eggs. Poultry Sci. 17:77.

Murphy, R. R., Hunter, J. E., and Knandel, H. C.: 1931, 1936. The vitamin D requirements of growing chicks and laying hens. Pa. Agr. Exper. Sta., Buls. 303 and 334.

Mussehl, F. E., and Ackerson, C. W.: 1935. Calcium and phosphorus requirements of growing turkeys. Poultry Sci. 11:147.

———, Hill, R. S., Blish, M. J., and Ackerson, C. W.: 1930. Utilization of calcium by the growing chick. Jour. Agr. Res. 40:191.

Norris, L. C., Heuser, G. F., Ringrose, A. T., and Wilgus, Jr., H. S.: 1934. Studies of the calcium requirement of laying hens. Poultry Sci. 13:308.

———, Heuser, G. F., Wilgus, Jr., H. S., and Ringrose, A. T.: 1933. The calcium and phosphorus requirements of laying hens. Cornell 46th Annual Rep. P. 137.

Patterson, F. D.: 1928. Gout in poultry. Vet. Med. 23:73.

Patton, A. R.: 1939. A study of glycine toxicity. Poultry Sci. 18:31.

———, Wilgus, Jr., H. S., and Harshfield, G. S.: 1939. The production of goiter in chickens. Science 89:162.

Poley, W. E., and Moxon, A. L.: 1938. Tolerance levels of seleniferous grains in laying rations. Poultry Sci. 17:72.

Prentice, J. H.: 1933a. The role of salt in poultry nutrition. I. Salt in the nutrition of the chick. Jour. Ministry Agr. Northern Ireland 4:72.

———: 1933b. The role of salt in poultry nutrition. II. Salt in the nutrition of the laying hen. Jour. Ministry Agr. Northern Ireland 4:92.

Quigley, G. D., and Waite, R. H.: 1932. Salt tolerance of baby chicks. Md. Agr. Exper. Sta., Bul. 340:313.

Rosen, F., Huff, J. W., and Perlzweig, W. A.: 1946. The effect of tryptophane on the synthesis of nicotinic acid in the rat. Jour. Biol. Chem. 163:343.

Russell, W. C.: 1939. The fat requirement of poultry. Abst. Rep. Cornell Nutr. Conf., Ithaca, N. Y.

———, Taylor, M. W., Walker, H. A., and Polskin, L. J.: 1942. The absorption and retention of carotene and vitamin A by hens on normal and low fat rations. Jour. Nutr. 24:199.

Schaible, P. J., Bandemer, S. L., and Davidson, J. A.: 1938. The manganese content of feedstuffs and its relation to poultry nutrition. Mich. Agr. Exper. Sta., Bul. T159.

———, Moore, J. M., and Conolly, R. A.: 1933. Factors influencing the incidence of perosis in Barred Rock chicks. Poultry Sci. 12:321.

Schlotthauer, C. F., and Bollman, J. L.: 1934. Experimental gout in turkeys. Proc. Staff Meetings Mayo Clin. 9:560.

Scott, H. M., Matterson, L. D., and Singsen, E. P.: 1947. Nutritional factors influencing growth and efficiency of feed utilization. I. The effect of the source of carbohydrate. Abst. Papers 36th Meet. Poultry Sci. Assn. P. 24.

Singsen, E. P.: 1947. Nutritional factors influencing growth and efficiency of feed utilization. II. The effect of protein level. Abst. Papers 36th Meet. Poultry Sci. Assn. P. 25.

———, Matterson, L. D., and Scott, H. M.: 1947. Phosphorus in poultry nutrition. III. The relationship between the source of vitamin D and the utilization of cereal phosphorus by the poult. Jour. Nutr. 33:13.

Sjollema, B.: 1935. Studies on the sodium requirement of chickens and on the consequences of a diet almost free of sodium. Tierernährung 7:184.

Smith, E. L.: 1939. Studies in the stability of vitamins A and D. II. Action of fatty peroxides on vitamin A. Biochem. Jour. 33:201.

Suffran, F.: 1909. Poisoning of poultry by salt (Trans. title). Rev. Gen. de méd. vét. 13:698.

Supplee, W. C.: 1935. A study of the effect of the significant variations of the calcium content of the A.O.A.C. basal rachitic ration on the percentage of bone ash in chick tibiae. Jour. Assn. Official Agr. Chem. 18:146.

Tepper, A. E., Durgin, R. C., and Charles, T. B.: 1939. Protein requirements of chickens at various stages of growth and development. N. H. Agr. Exper. Sta., Bul. 312.

Titus, H. W., Byerly, T. C., Ellis, N. R., and Nestler, R. B.: 1937. Effect of the calcium and phosphorus content of the diet of chickens on egg production and hatchability. Poultry Sci. 16:118.

Tomhave, A. E.: 1939. Protein levels of rations for White Leghorn pullets. Del. Agr. Exper. Sta., Bul. 219.

Torrey, J. P., and Graham, R.: 1935. A note on experimental salt poisoning in ducks. Cornell Vet. 25:50.

Tully, W. C., and Franke, K. W.: 1935. A new toxicant occurring naturally in certain samples of plant foodstuffs. VI. A study of the effect of affected grains on growing chicks. Poultry Sci. 14:280.

Watkins, W. E., and Mitchell, H. H.: 1936. The phosphorus requirements of growing chickens, with a demonstration of the value of controlled experimental feeding. Poultry Sci. 15:32.

Welch, H.: 1928. Goiter in farm animals. Mont. Agr. Exper. Sta., Bul. 214.

Wiese, A. C., Benham, G. H., Elvehjem, C. A., and Hart, E. B.: 1941. Further bone phosphatase studies in chick perosis. Poultry Sci. 20:255.

———, Johnson, B. C., Elvehjem, C. A., Hart, E. B., and Halpin, J. G.: 1939. A study of blood and bone phosphatase in chick perosis. Jour. Biol. Chem. 127:411.

Wilcox, J. S.: 1934. The nitrogen balance of laying hens. Jour. Agr. Sci. 24:636.

Wilder, O. H. M., Bethke, R. M., and Record, P. R.: 1933. The iodine content of hens' eggs as affected by the ration. Jour. Nutr. 6:407.

Wilgus, Jr., H. S.: 1931. The quantitative requirements of the growing chick for calcium and phosphorus. Poultry Sci. 10:107.

———, Gassner, F. X., Patton, A. R., and Gustavson, R. G.: 1941a. The goitrogenicity of soybeans. Jour. Nutr. 22:43.

———, Harshfield, G. S., Patton, A. R., Ferris, L. P., and Gassner, F. X.: 1941b. The iodine requirements of growing chickens. Poultry Sci. 20:477.

———, Norris, L. C., and Heuser, G. F.: 1936. The role of certain inorganic elements in the cause and prevention of perosis. Science 84:252.

———, Norris, L. C., and Heuser, G. F.: 1937a. The role of manganese and certain other trace elements in the prevention of perosis. Jour. Nutr. 14:155.

———, Norris, L. C., and Heuser, G. F.: 1937b. The effect of various calcium and phosphorus salts on the severity of perosis. Poultry Sci. 16:232.

Winter, A. R., Dakan, E. L., and Bayes, A.: 1932. Protein levels for finishing pullets. Poultry Sci. 11:30.

CHAPTER SEVEN

# VITAMINS AND VITAMIN DEFICIENCIES

*By* R. M. SHERWOOD AND J. R. COUCH, *Department of Poultry Husbandry,*
*Texas Agricultural Experiment Station, College Station, Texas*

★   ★   ★

Vitamins may be described as a group of organic substances which have definite biological effects when present in the diet in minute quantities. It is thought that in many cases their biological effects are results of catalytic action.

The term vitamins is a very modern one, dating back less than thirty-five years (Funk, 1912). However, symptoms of a disease, later found to be caused by a food deficiency, were recognized by the Chinese as early as 2600 B.C. (Harris, 1938). This condition is now known as beriberi and is caused by a deficiency of vitamin $B_1$.

According to Hippocrates, who lived from 460–359 B.C., certain foods in the diet prevented night blindness (McCollum *et al.*, 1939). He advocated an abundance of liver as a remedy for night blindness. His recommendations are in accord with our present concepts of vitamin A as a nutritional entity. Later, Lind (1753) stated that fresh fruits and vegetables would definitely prevent and cure scurvy, a deficiency disease due to lack of vitamin C or ascorbic acid.

Notwithstanding these early observations, no basic experimental work was done with purified food materials until Lunin (1881), a Swiss investigator, fed synthetic diets and concluded that animals could not survive on diets made up wholly of purified protein, carbohydrates, fats, and salts.

By the use of synthetic diets, Eijkman (1897), in Java, in 1890, succeeded in producing experimental polyneuritis in fowls (Grijns, 1935). Holst and Frölich (1907), in Norway, attempted to produce polyneuritis in guinea pigs by the use of unbalanced diets, but experimental scurvy resulted.

Funk (1912), a Polish biochemist, after having attempted to isolate the beriberi preventive factor, suggested the term "vitamines" as a name for all accessory food factors. McCollum and Davis (1913, 1915) presented the first evidence of the existence of both water-soluble and fat-soluble substances.

From 1915 to the present time, work has progressed rapidly, and many reports have been published. Many vitamins have been isolated, and their structures have been established.

Emslie (1934), Cruickshank (1935), and Jukes (1937) are almost unanimous in the belief that vitamins A, D, and G (riboflavin) are the ones that are most frequently deficient in poultry diets. Under conditions where rapid gains in weight or high egg production are obtained, the requirements of all food materials are more exacting than when poorer results are secured. Under these conditions, it is possible that other vitamins may be deficient in diets not thoroughly balanced.

In preparing this chapter, no effort was made to list in the bibliography all the papers on the deficiency diseases in question. The literature selected gives a rather complete description of the particular deficiency disease discussed. The student may, by the use of the bibliographies given, extend his knowledge on a particular deficiency disease.

### GENERAL REFERENCES

Cruickshank, E. M.: 1935. Vitamins and minerals in poultry nutrition. Nutr. Abst. and Rev. 5:1.

Eijkman, C.: 1897. Eine Beri Beri-ähnliche Krankheit der Hühner. (Virchow's) Arch. f. Path. Anat. 148:523.

Emslie, A. R. G.: 1934. Recent research in poultry nutrition. Imp. Bur. of Animal Nutr., Tech. Com. No. 5, Rowett Institute, Aberdeen, Scotland.

Funk, C.: 1912. The etiology of the deficiency diseases. Jour. State Med. 20:341.

Grijns, G.: 1935. Researches on vitamins, 1900–1911. Gorinchem. J. Noorduyn en Zoon N. V.

Harris, L. J.: 1938. Vitamins and Vitamin Deficiencies, Vol. 1. Introductory and historical. Vitamin $B_1$ and beri-beri. P. Blakiston's Son and Co., Inc., Philadelphia, Pa.

Holst, A., and Frölich, T.: 1907. Experimental studies relating to "Ship Beriberi" and scurvy. II. On the etiology of scurvy. Jour. Hyg. 7:634.

Jukes, T. H.: 1937. Recent studies of vitamins required by chicks. Jour. Nutr. 13:359.

Lind, J.: 1753. A Treatise on Scurvy. Sands, Murray, and Cochrane, London.

Lunin, N.: 1881. Ueber die Bedeutung der anorganischen Salze für die Ernährung des Thieres. Zeitschr. Physiol. Chem. 5:31.

McCollum, E. V., and Davis, M.: 1913. The necessity of certain lipins in the diet during growth. Jour. Biol. Chem. 15:167.

———— and Davis, M.: 1915. The nature of the dietary deficiencies of rice. Jour. Biol. Chem. 23:181.

————, Orent-Keiles, E., and Day, H. G.: 1939. The Newer Knowledge of Nutrition. Fifth ed. The Macmillan Co. New York.

The Vitamins. 1939. A symposium arranged under the auspices of the Council on Pharmacy and Chemistry and the Council on Foods of the Am. Med. Assn., Am. Med. Assn., Chicago, Ill.

### VITAMIN A AND VITAMIN A DEFICIENCY

Vitamin A is essential in poultry for the support of life and growth. Beach (1924), Seifried (1930), and Cruickshank (1935) report that it is also necessary for maintaining the normal structure of the epithelial lining of the respiratory and upper alimentary tracts. Vitamin A has an empirical formula of $C_{20}H_{30}O$. It is an unsaturated primary alcohol, containing one beta ionone ring and five double bonds one of which is localized in this ring. The beta ionone ring is very important biologically according to Palmer (1939), if it is altered in any respect the compound no longer exerts vitamin A activity in the animal body. Vitamin A occurs in the animal body and in fish liver oils. Palmer (1939) and Peterson et al. (1939) report that vitamin A is not found in plants, but its precursors, carotenes and xanthophyll, are found in green plants and yellow corn. Palmer (1939) also reports

that it is the only vitamin known that is produced by animal metabolism of precursors, which are the products of plant metabolism. He also reports that there are three physiologically important carotenes; these are alpha, beta, and gamma carotene. The carotenes are unsaturated hydrocarbons; the number of molecules of vitamin A, which may be formed in the animal body from a molecule of these carotenes, is dependent upon the number of unaltered beta ionone rings which they contain. A molecule of beta carotene has two such rings, and will form two molecules of vitamin A in the animal body. Beta carotene is much more abundant in carotene-containing feeds than alpha or gamma carotene. A molecule of either alpha or gamma carotene has only one beta ionone ring, and only one molecule of vitamin A may be formed from each of these precursors. Yellow corn contains cryptoxanthin, which has one unaltered beta ionone ring and one beta ionone ring having a hydroxyl group in place of one hydrogen, thereby forming a secondary alcohol; aside from this, the constitution of cryptoxanthin is the same as that of beta carotene (Palmer, 1939; Peterson *et al.*, 1939). Cryptoxanthin will form only one molecule of vitamin A, since one of its beta ionone rings has been altered to the extent mentioned above. There is probably some biological significance attached to the primary alcohol groups of the vitamin A molecule. Evidence exists to indicate that vitamin A is absorbed and transported as an ester with fatty acids, proteins, or bile acids. There is also good evidence, reported by Palmer (1939) and Bessey and Wolbach (1939), that the factor which is essential for the prevention of night blindness is a protein-vitamin A compound in the body.

### CLINICAL SYMPTOMS

Vitamin A deficiency in chicks is characterized by a cessation of growth at from three to four weeks of age, by drowsiness, weakness, incoordination, staggering gait, emaciation, and ruffled plumage (Cruickshank, 1935; Emmett and Peacock, 1923; Cruickshank *et al.*, 1927; Elvehjem and Neu, 1932; Miller and Bearse, 1934; Tepper and Durgin, 1938; Sherwood, 1939). There is a lack of yellow pigment in the shanks and beaks in breeds of chickens that usually have this pigment, and the combs and wattles of the chicks are usually pale. In some cases there is lachrymation and the presence of cheesy-like material under the eyelids. Xerophthalmia is a definite symptom of vitamin A deficiency, but according to Cruickshank (1935), all chicks do not exhibit this symptom, because they often die before the eyes become affected.

Adult birds with vitamin A deficiency are emaciated and weak, and their feathers are ruffled (Beach, 1924). According to Sherwood and Fraps (1932) there is a marked decrease in egg production, and the length of time between clutches increases greatly. Polk and Sipes (1940) report that the deficiency

causes a very great decrease in hatchability of the eggs and an increase in embryonic mortality in eggs from affected birds. A watery discharge from the nostrils and eyes is noted; the eyelids are often glued together (Beach, 1924). As the deficiency continues, an accumulation of milky white, caseous material forms in the eyes. In the latter stages of the disease, the eyes become filled with this white exudate to such an extent'that it is impossible for the bird to see unless the mass is removed; in many cases the eye is destroyed (Beach, 1924; Sherwood, 1939).

According to Sherwood and Fraps (1936) the length of time that chicks can survive on diets deficient in vitamin A depends upon the feed consumed by the "mother hen." It is reported by Cruickshank (1935) and Sherwood and Fraps (1936) that if feed is very low in vitamin A, the mortality of her chicks will be high during the first two weeks of their life regardless of the amount of vitamin A in the chick diet. If the "mother hen" was fed an adequate amount of vitamin A, but the chicks are fed an inadequate amount of this vitamin, the survival period of the chicks will extend from four to seven weeks.

The length of time a hen can survive on a vitamin A deficient diet depends upon the amount of vitamin A stored in the liver and other tissues of her body, the state of growth and development of the hen, and the number of eggs she lays (Sherwood and Fraps, 1932). Hens which previously have been fed adequate amounts of vitamin A usually die within two to five months after being placed on a diet practically devoid of this vitamin or its precursors.

### PATHOLOGY

*Gross lesions* that are apparent on post-mortem examination of chickens affected with vitamin A deficiency are described by Beach (1924), Cruickshank (1935), Emmett and Peacock (1923), Elvehjem and Neu (1932), and Sherwood (1939) as being confined to the mucous membranes of the head, esophagus, crop, and respiratory tract. Nodular, pustule-like lesions are numerous in the affected areas, and these vary greatly in different individuals. Aside from the involvement of the respiratory and upper alimentary tracts, the kidneys are the only other organs which show consistent definite lesions in vitamin A deficiency. They become pale and show a network of fine, white lines, which are the renal tubules filled with urates. In extreme cases, even the ureters are filled with urates. Deposits of urates have also been found on the heart, pericardium, omentum, liver, and spleen of affected birds. According to Elvehjem and Neu (1932) the urates result from pathological changes in the kidney due to vitamin A deficiency which prevent the normal elimination of uric acid. This is accompanied by an increase in the uric acid content of the blood of affected birds.

*Upper respiratory tract.* The clinical symptoms and pathological lesions of vitamin A deficiency of the respiratory tract are variable, and it is difficult to differentiate this condition from infectious coryza, virus-diphtheria, and infectious tracheal bronchitis (Beach, 1924; Seifried, 1930). In the case of infectious coryza and virus-diphtheria, an exudate develops in the nasal passages and sinuses which later is transformed into white or slightly yellowish, caseous masses; these, at times, completely plug the cleft palate. With infectious coryza and virus-diphtheria, thin membranes also appear which, together with the nasal plug, may easily be confused with the lesions of vitamin A deficiency. In the latter condition these membranes may also be present but are usually limited to the cleft palate and its adjacent epithelium and may easily be removed without bleeding. According to Seifried (1930) this is not true in virus-diphtheria.

There are rather definite tissue changes in the respiratory tract of chickens affected with vitamin A deficiency. There is atrophy and degeneration of the respiratory mucous membrane and its glands. Later the original epithelium is replaced by a stratified squamous, keratinizing epithelium (scalelike lining). In the early stages of vitamin A deficiency in chickens, the turbinates are filled with seromucoid water-clear masses, which may be forced out of the nodules and cleft palate by the application of slight pressure. Early in the disease, the vestibule becomes plugged and overflows into the paranasal sinuses; this is due to the consistency of the exudate and the complicated structure of the nasal fossae. The exudate may also be forced through the cleft palate and in this case fills up with white or slightly yellow, caseous masses the region around the vomer, which includes the sinuses and other nasal cavities. This causes a swelling of one or both sides of the face.

After the sinuses have filled, the tear ducts become occluded. The eyeball is pressed against the frontal bones and is sometimes forced out to the side because it cannot move in a ventral direction. Upon removal of the inflammatory products, the mucous membranes of the nasal passages, sinuses, mouth, and throat appear thin, rough, and dry. Unattached masses of caseous material often form in the cleft palate and in the mucous membranes of the roof of the mouth.

Lesions in the larynx and trachea of vitamin A deficient chickens occur both in the early and also in the later stages of the disease. Near the entrance of the pharynx, the lesions consist of pustule-like patches of white, caseous material. Caseous, crumbly, white masses often appear in the mucous membrane of the anterior end of the larynx on its ventral side and in the pointed angle which is formed by the cartilage of the larynx. Frequently, similar lesions may be found in the trachea and bronchi. In the early stages these may be difficult to see. As the condition progresses, the mucous membrane is covered with a dry, dull, fine film, which is slightly uneven, whereas the

normal membrane is even and moist. In some cases, small, nodule-like particles are in or beneath the mucous membrane in the upper part of the trachea. These lesions are much more striking in the latter stages of the
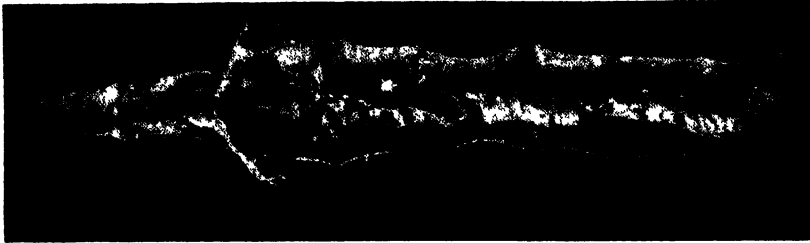


FIG. 7.1. Trachea of chicken with A avitaminosis showing desquamated epithelium partly in the form of a tube. Bird died after 87 days on experimental diet. (Seifried, Jour. Exper. Med.)

deficiency and may then be seen easily with the naked eye. The formation of a thin membranous covering over the mucosa of the trachea and bronchi is a symptom of vitamin A deficiency but is often mistaken for that of infectious tracheitis (Fig. 7.1). The smaller bronchi often become completely occluded with these membranes. These changes vary in individual birds; in some cases the larynx shows the most marked changes, while in other cases the trachea does.

*Upper alimentary tract.* Seifried (1930) points out that small, white, pustule-like lesions are found in the mouth, esophagus, and pharynx and may



FIG. 7.2. Pustule-like lesions in pharynx and esophagus—vitamin A deficiency. (Biester and Schwarte, No. Am. Vet.)

extend into the crop. Small, white, or slightly yellowish, pustule-like lesions from five-tenths to 2 mm. in diameter appear in the region of the excretory ducts of the glands of the mouth quite early in vitamin A deficiency in chickens. These lesions are raised above the surface of the mucous membrane but show a depression in the center. Small ulcers, surrounded by inflammatory products, may appear at the site of these lesions. This condition very closely resembles that of fowl pox; it is doubtful if a differential diagnosis can be made without microscopic examination. The watery exudate in the nasal cavity gradually becomes transformed into white or slightly yellowish, caseous masses, and at times the cleft palate is completely plugged. Pustule-like patches are present on the mucous membrane of the esophagus and may extend into the posterior part of the crop where the folds of the esophagus are continued (Fig. 7.2). No gross lesions have been found in the intestines.

## HISTOPATHOLOGY

*Respiratory tract.* The first lesion which appears is an atrophy of the cytoplasm and a loss of the cilia in the columnar ciliated epithelium (Seifried, 1930). The nuclei often present a marked karyorrhexis. A pseudomembrane, formed by the atrophying and degenerating ciliated cells, may hang like tufts on the basement membrane; later these are pushed off. During this process, new cylindrical or polygonal cells may be formed either singularly or in pairs and appear as islands beneath the epithelium. These new cells become more and more numerous, and their nuclei become larger and contain less chromatin. The cell boundaries become less clearly defined; finally, the columnar ciliated epithelial lining of the trachea, bronchi, and submucous glands becomes transformed into a squamous, stratified, keratinizing epithelium (Fig. 7.3). Seifried (1930) concludes that this process is not related to bacterial infection.

The nasal cavities and communicating sinuses show essentially the same epithelial lesions as the trachea. All parts of the nasal cavities are generally involved. There is an increased proliferation of the superficial epithelial cells in the gland-free part of the nasal vestibule, regiovestibularis; the alteration is not a real keratinization. A true keratinization, similar to that found in the trachea, appears in the mucous membrane of the roof of the nasal vestibule, particularly in the median and dorsal part of the concha of the vestibule (Fig. 7.4). Seifried (1930) is of the opinion that the epithelium of the glands becomes involved somewhat later than the epithelium of the mucous membrane. The mucous membrane of the paranasal sinuses is thin, and only a few glands are found on the median wall. Keratinization occurs in the excretory duct of the lateral nasal gland, Glandula lateralio nasi, and in the nasolacrimal duct. The main lesion of the lateral nasal gland is in the ducts and collecting spaces.

*Upper alimentary tract and associated glands.* Lesions in the glands of this region are very much the same as those found and described above in the respiratory tract (Seifried, 1930). Some peculiarities are, however, ap-



FIG. 7.3. Cross section through trachea showing newly formed stratified epithelium. Several cells near surface showing "balloon" degeneration. ×990. (Seifried, Jour. Exper. Med.)

parent. The early lesions of these glands are found in the collecting spaces and ducts and may appear earlier in the maxillary and submaxillary glands than in the nasal passages, sinuses, trachea, or bronchi. As the disease progresses, the collecting spaces become filled with masses of mucus, degenerated cells, and inflammatory products. These accumulations are



FIG. 7.4. Nasal septum. Complete replacement of surface epithelium by stratified keratinized epithelium. ×50. (Seifried, Jour. Exper. Med.)

probably due to lesions in the excretory ducts of the glands. The epithelium of the mucous membrane of the mouth extends into the duct, which has been partially filled with the stratified, keratinized epithelium, and more or less complete occlusion results (Fig. 7.5). The glands, in the early stages, continue to secrete mucus, which accumulates in the collecting spaces. Desquamated cells from the newly formed stratified epithelium become more and
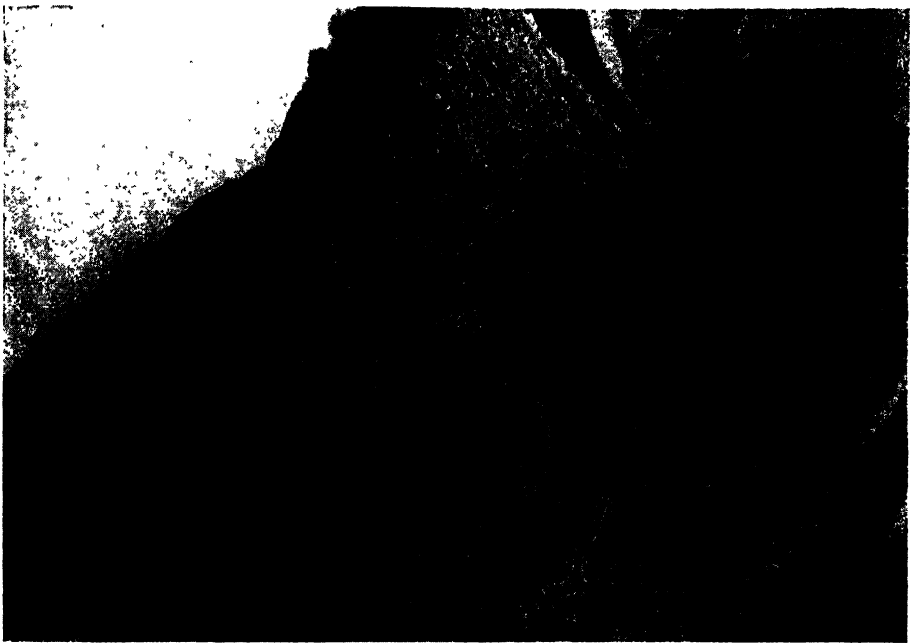


FIG. 7.5. Cross section through base of tongue showing early keratinization and degeneration of the upper layers of the newly formed epithelium. ×80. (Seifried, Jour. Exper. Med.)

more numerous, the glands becoming smooth and distended, although originally they were sacs with invaginations. This distended sac finally becomes completely filled with stratified keratinized epithelial cells (Fig. 7.6). The above applies principally to the tongue, palate, and esophagus. Seifried (1930) believes that these lesions, resulting from the lack of vitamin A, may enable bacteria to enter the body. Such bacterial infections are more prevalent in the mouth cavity than in the crop and esophagus.

### FEEDING RECOMMENDATIONS

The first four to six weeks of a chick's life is a critical period from the standpoint of vitamin A metabolism, because during this period, the vitamin A storage increases very slowly even on a well-fortified diet (Rubin and Bird, 1941). This is especially important if the egg from which the chick hatched

was produced by a hen receiving a diet low in vitamin A (Cruickshank, 1935; Sherwood and Fraps, 1936).

Fish liver oil is an important source of vitamin A for poultry. Fortified fish liver oils, which contain not less than 3,000 international units of vitamin A per gram are most satisfactory. Such oils will not flavor the flesh or the eggs when fed in quantities sufficient to completely protect the birds against vitamin A deficiency.

It is fortunate that the animal can produce vitamin A from carotene and



FIG. 7.6. Cross section through base of tongue showing final stage of process with dilatation of the glands which are filled with stratified, more or less keratinized, homogeneous masses. ×50. (Seifried, Jour. Exper. Med.)

cryptoxanthin. Carotene is found in choice green feeds, such as lawn clippings, short succulent pasture, and dehydrated alfalfa leaf meal, and in carrots, sweet potatoes, and crystalline carotene in oil; cryptoxanthin is found in yellow corn. Palmer (1939) and Peterson et al. (1939) report that a molecule of cryptoxanthin produces only one-half as much vitamin A as a molecule of beta carotene. It should be possible to provide all of the vitamin A requirements of poultry by feeding choice dehydrated alfalfa leaf meal or fresh green feed (Sherwood, 1939; Sherwood and Fraps, 1932; Sherwood and Fraps, 1936). It is essential that the dehydrated alfalfa leaf meal or green feed be of good quality. A rich green color, in the absence of brown pigment, is a reasonably good indicator of good quality dehydrated alfalfa leaf meal.

Fraps and Kemmerer (1937) have pointed out that vitamin A and carotene are destroyed in the feed if it is exposed to the air. Losses in this case are greater at high temperatures than in cold weather and are greater when

the vitamin A or carotene containing feeds are mixed with other feeds. Mixed feeds should be fed as soon as possible after mixing. Increasing amounts of carotene oxidase and fat in the individual feeds increase the losses of carotene and vitamin A in mixed feeds (Sumner and Dounce, 1939; Sumner and Sumner, 1940; Sherwood and Fraps, 1940). Samples of different lots of individual feeds stored under different conditions vary widely in their carotene oxidase content (Sherwood and Fraps, 1940). It is not known whether such differences are present in the individual feeds before storage or whether they are the result of storage conditions. Because of the many factors which change or destroy the carotene and vitamin A of feeds, mixed feeds that are thought to contain adequate amounts of carotene and vitamin A may sometimes actually be deficient in this vitamin or its precursors when fed.

Different laboratories are not in complete agreement concerning the amount of vitamin A required by different classes of poultry (Cruickshank et al., 1927; Sherwood, 1939; Almquist and Mecchi, 1939). In Table 1

TABLE 1
VITAMIN A REQUIREMENTS OF DIFFERENT CLASSES OF POULTRY PER 100 POUNDS OF FEED

|  | Recommended International Units of Vitamin A |
|---|---|
| Chicks.. ..... . . . .. .. . | 200,000 |
| Growing pullets.. . .. . .. | 200,000 |
| Laying hens..... .. . .. | 330,000 |
| Breeding hens.. .... . | 330,000 |
| Starting poults .... . . .. .. ... .. | 400,000 |
| Growing turkeys... .. .. | 400,000 |
| Breeding turkeys.... . . . | 400,000 |

amounts of vitamin A recommended by the Subcommittee on Poultry Nutrition of the National Research Council (1946) are given.

In order to calculate how much choice dehydrated alfalfa leaf meal is required in 100 pounds of feed, one must first know how much carotene the alfalfa leaf meal contains. For example, alfalfa leaf meal may be assumed to contain 50,000 micrograms of carotene per pound. Since 1 microgram is equivalent to approximately 1⅔ international units, this would mean that a pound of this meal would contain about 85,000 international units of vitamin A per pound. In calculating a chick diet which is to contain 135,000 international units per 100 pounds, it would require 1.6 pounds (135,000 divided by 85,000) of meal per 100 pounds of chick feed. It is not uncommon to consider a safety factor of 2 to allow for poorer alfalfa leaf meal and for the oxidation of carotene; in that case, the 1.6 pounds is multiplied by 2, which gives approximately 3 pounds of alfalfa leaf meal required per 100 pounds of chick feed.

## TREATMENT OF SEVERE CASES OF VITAMIN A DEFICIENCY

Poultry that has been receiving a ration low in vitamin A content should be given, for a few weeks, higher amounts of this vitamin than those recommended in the above table. Doses as high as one-half of 1 per cent of fortified fish liver oil can be recommended under these conditions. It is well to mix the fish liver oil with a small amount of mash *daily* and feed it as a moist mash. If this procedure is followed, the loss of vitamin A will be negligible. Turkeys should be given twice as large a dose of fish liver oil as chickens (Hinshaw and Lloyd, 1934).

According to Bessey and Wolbach (1939) absorption of vitamin A in the body reaches a maximum in less than one day after it is fed. For that reason, birds that are not in advanced stages of vitamin A deficiency respond promptly. Sherwood and Fraps (1940) found that hens that are in rather advanced stages of deficiency have, in some instances, returned to production in less than a month after the diet was corrected, while others die notwithstanding treatment.

## REFERENCES ON VITAMIN A

Almquist, H. J., and Mecchi, E.: 1939. Vitamin A requirements of laying hens. Poultry Sci. 18:129.
Beach, J. R.: 1924. Studies on a nutritional disease of poultry caused by vitamin A deficiency. Calif. Agr. Exper. Sta., Bul. 378.
Bessey, O. A., and Wolbach, S. B.: 1939. Vitamin A physiology and pathology. The Vitamins, Am. Med. Assn., Chicago, Ill. P. 27.
Cruickshank, E. M.: 1935. Vitamins and minerals in poultry nutrition. Nutr. Abst. and Rev. 5:1.
———, Hart, E. B., and Halpin, J. G.: 1927. The vitamin A and vitamin D content of cod liver meal. Poultry Sci. 7:9.
Elvehjem, C. A., and Neu, V. F.: 1932. Studies in vitamin A avitaminosis in the chick. Jour. Biol. Chem. 97:71.
Emmett, A. D., and Peacock, G.: 1923. Does the chick require the fat-soluble vitamins? Jour. Biol. Chem. 56:679.
Fraps, G. S., and Kemmerer, A. R.: 1937. Losses of vitamin A and carotene from feeds during storage. Tex. Agr. Exper. Sta., Bul. 557.
Hinshaw, W. R., and Lloyd, W. E.: 1934. Vitamin-A deficiency in turkeys. Hilgardia 8:281.
Miller, M. W., and Bearse, G. E.: 1934. A comparison of some vitamin A supplements for chick feeding. Wash. Agr. Exper. Sta., Bul. 292.
National Research Council: 1946. Recommended nutrient allowances for poultry. Mimeo. data No. 1.
Palmer, L. S.: 1939. The chemistry of vitamin A and substances having a vitamin A effect. The Vitamins, Am. Med. Assn., Chicago, Ill. P. 15.
Peterson, W. J., Hughes, J. S., and Payne, L. F.: 1939. The carotenoid pigments: occurrence, properties, methods of determination, and metabolism by the hen. Kan. Agr. Exper. Sta., Tech. Bul. 46.
Polk, H. D., and Sipe, G. R.: 1940. The effect of vitamin A deficiency on malposition of the chick embryo. Poultry Sci. 19:396.
Rubin, M., and Bird, H. R.: 1941. Some experiments on the physiology of vitamin A storage in the chick. Poultry Sci. 20:291.
Seifried, O.: 1930a. Studies on A-avitaminosis in chickens. Lesions of the respiratory tract and their relation to some infectious diseases. Jour. Exper. Med. 52:519.
———: 1930b. Lesions of the upper alimentary tract and their relation to some infectious diseases. Jour. Exper. Med. 52:533.
Sherwood, R. M.: 1939. Vitamin A requirements of poultry. Proc. Seventh World's Poultry Cong. P. 123.
——— and Fraps, G. S.: 1932. The quantities of vitamin A required by pullets for maintenance and for egg production. Tex. Agr. Exper. Sta., Bul. 468.
——— and Fraps, G. S.: 1936. The quantities of vitamin A required by growing chicks. Tex. Agr. Exper. Sta., Bul. 528.
——— and Fraps, G. S.: 1940. Unpublished data.
Sumner, J. B., and Dounce, A. L.: 1939. Carotene oxidase. Enzymologia 7:130.

——— and Sumner, R. J.: 1940. The coupled oxidation of carotene and fat by carotene oxidase. Jour. Biol. Chem. 134:531.

Taylor, M. W., and Russell, W. C.: 1947. The provitamin A requirements of growing chickens. Poultry Sci. 26:234.

———, Stern, J. R., Russell, W. C., and Jungherr, E.: 1947. The provitamin A requirements of hens. Poultry Sci. 26:243.

Tepper, A. E., and Durgin, R. C.: 1938. Vitamin-A requirements of growing chicks—a review of experimental work. N. H. Agr. Exper. Sta., Bul. 310.

## VITAMIN D AND VITAMIN D DEFICIENCY

The term vitamin D is used to designate at least ten different sterol derivatives all of which exhibit antirachitic properties (Bills, 1939). These different forms of the vitamin do not include the antirachitic vitamins of fish oils, which to date have comprised the most extensive source of vitamin D available to the poultryman. Ergosterol, which is found in yeast, is a sterol that acquires vitamin D activity through absorption of ultraviolet light and in the process is converted to calciferol (Bills, 1939). This is known as vitamin $D_2$, but it is not a satisfactory antirachitic vitamin for chickens.

Cholesterol, an animal sterol, contains 7-dehydro-cholesterol, which likewise acquires vitamin D activity on absorption of ultraviolet light (Bills, 1939). It is known as vitamin $D_3$ and, according to Koch and Koch (1941), is the vitamin which is formed in the skin and fur or feathers of animals upon exposure to sunlight or other sources of ultraviolet rays. It was shown by Hart et al. (1923) that sunlight was important in the rearing of baby chicks. There is evidence that vitamin $D_3$ is the principal form found in fish-liver oil. Irradiated 7-dehydro-cholesterol or vitamin $D_3$ is an excellent source of vitamin D for chickens (Bills, 1939).

Bone development of the chicken is pointed out by Fell (1925) to be similar, if not identical, to that of the mammal. Evidence has been presented by Nonidez (1928) and McGowan and Emslie (1934) to show that a deficiency of the antirachitic vitamin in the diet of the chicken brings about a disorder showing all of the essential characteristics of mammalian rickets. Rickets is due to a deficiency of mineral material in the bones. The relationship of vitamin D to the decrease or increase of calcium and phosphorus in the bone or total bone ash is somewhat of a disputed question (McGowan and Emslie, 1934; Liu, 1940; Smith and Spector, 1940; Nicolaysen, 1937a, 1937b, 1937c; Innes and Nicolaysen, 1937). Shipley (1924) states that rachitic bones calcify in vitro when placed in normal serum or plasma. There is evidence which indicates that vitamin D has an influence on the architecture of the bones. Vitamin D promotes the absorption of calcium from the intestinal tract, but does not exert such an influence on phosphorus. It also tends to prevent the excretion of calcium.

### SYMPTOMS OF VITAMIN D DEFICIENCY IN CHICKS

The first striking symptom of vitamin D deficiency with chicks is a condition which was termed leg weakness by the early workers (Hart et al., 1922;

Pappenheimer and Dunn, 1925; Drummond 1916). Maughan (1928) states that leg weakness is followed by retardation of growth, roughened feathers, and general unthriftiness (Fig. 7.7). Affected birds exhibit a halting gait and tenderness of the joints as noted by the position of the feet. The birds often die of starvation if feed and water are not easily accessible. Growth is normal at first in chicks fed a diet deficient in the antirachitic vitamin from hatching time, but later ceases. The bones become soft, and marked skeletal distortions become apparent (Hart et al., 1922; Goodale, 1926; Nonidez and Goodale, 1927; Hughes et al., 1925a; Doyle, 1940). The deformities resulting from a deficiency of vitamin D are found principally in the legs and at the junction of the ribs on the sides of the breast. The spinal column may bend down, resulting in a downward abnormal curve in the sacral and coccygeal region. The sternum usually has an acute dent or bend somewhere near its center. This reduces the size of the thorax, with the consequent crowding of the vital organs. A characteristic beading of the ribs results in a row of



FIG. 7.7.  Birds showing rickets.

distinctive enlargements at the junctions of the dorsal and ventral portions and at the articulation of the ribs with the vertebrae. This further reduces the size of the thoracic cavity (Maughan, 1928). The claws and upper mandible become soft and much longer than those found on normal birds.

Steenbock et al. (1923) and Ackerson et al. (1925) state that in addition to the bone deformities observed in rachitic chickens, the calcium and phosphorus contents of the blood are lower than normal. The phosphatase concentration of the blood of such birds is higher than normal (Correll and Wise, 1938). Nonidez and Goodale (1927) state that the parathyroid glands of rachitic chickens hypertrophy.

### SYMPTOMS OF VITAMIN D DEFICIENCY IN MATURE FOWL

Doyle (1925) and Hughes and Payne (1924), point out that rickets develops in mature birds about two to three months after they are deprived of vitamin D. An increase in the number of thin-shelled and soft-shelled eggs, which the hens lay, is an indication of a deficient intake of vitamin D. Some hens lose the use of their legs temporarily but recover this function after laying an egg (Doyle, 1925). The beak often becomes soft and pliable.

Ribs of affected birds lose their normal rigidity and turn inward at the junction of the sternal and vertebral portions. This produces an inward curve of the ribs along the side of the thorax. The sternum is usually bent. The calcium, phosphorus, and vitamin D content of eggs from rachitic hens is lower than in eggs from hens fed non-rachitic diets. According to Hughes and Payne (1924), Hughes et al. (1925b), and Hart et al. (1925), hatchability of eggs from hens affected with vitamin D deficiency is much lower than those of hens receiving an adequate amount of this vitamin.

### PATHOLOGY OF VITAMIN D DEFICIENCY IN MATURE BIRDS

In fowls receiving a deficient amount of the antirachitic vitamin, the characteristic changes observed on post-mortem are confined to the bones and parathyroid glands. The bones are soft and break easily. Well-defined knobs are present on the inner surface of the ribs where the sternal portions join the vertebral portions. Many of the ribs have undergone a spontaneous fracture in the region where the knobs are located. Large callouses are present at the points of fracture. Skeletal changes also appear in the vertebral column, pelvis, and sternum. Histological sections of the leg bones show a deficiency of calcium and an excess of osteoid tissue.

### RECOMMENDATIONS

Vitamin D is required by chickens and other fowls when they do not have access to direct sunlight. Some fish-liver oils and dry irradiated products have been shown to be satisfactory sources of vitamin D for chickens. The fish-liver oils contain vitamins A and D, while many of the dry irradiated products on the market contain only vitamin D. Sunlight is important in the rearing of baby chicks and acts as a supplement or equivalent to the anti-rachitic factor. Extensive research was necessary to show that ultraviolet light is as active as sunlight in synthesizing vitamin D in the skin of birds.

Growing chicks should receive approximately 180 A.O.A.C.[1] chick units of vitamin D per pound of diet. This is approximately twice the amount of vitamin D necessary to meet the minimum requirements of growing chicks (Couch et al., 1935; Murphy et al., 1936; Carver et al., 1934). Laying hens should receive approximately 454 A.O.A.C. chick units of vitamin D per pound of diet. This is also somewhat higher than that found to meet the minimum requirements of laying hens.

It is necessary to ascertain the vitamin D content of the fish-liver oil or dry irradiated product to be used, and then figure the amount needed to fill the recommendations given.

Ducklings are thought to require about the same amount of vitamin D as chickens (Fritz et al., 1941).

---

[1] Association of Official Agricultural Chemists.

There are wide variations in the recommendations of the amount of vitamin D required for turkeys. They range from 375 to 900 A.O.A.C. chick units per pound of feed. The growth of poults raised on a low vitamin D diet was in proportion to the level of vitamin D in the diet of the breeding hens. Breeding turkeys, for egg production and hatchability, should receive 800 A.O.A.C. chick units of vitamin D per pound of diet. Increasing amounts of vitamin D up to 1,800 A.O.A.C. units per pound resulted in increased weight of eggs and in decreased numbers of soft-shelled eggs (Wilhelm *et al.*, 1941). According to Sloan (1934), a seasonal variation exists in the antirachitic effectiveness of sunshine. Sloan (1934) and Couch *et al.* (1939) report that chickens must be exposed to direct sunlight 30 minutes per day in the winter, 5 to 10 minutes per day in the spring, and 2½ minutes per day in the summer in order to meet their vitamin D requirements. According to the revised recommendations of the poultry subcommittee of the National Research Council, baby chicks require 180 A.O.A.C. units, growing chicks 180, and hens 450 per pound of feed. For turkeys they recommend 800 A.O.A.C. units per pound of feed for all ages.

## REFERENCES ON VITAMIN D

Ackerson, C. W., Blish, M. J., and Mussehl, F. E.: 1925. A study of the phosphorus, calcium, and alkaline reserve of the blood sera of normal and rachitic chicks. Jour. Biol. Chem. 63:75.

Bills, C. E.: 1939. The chemistry of vitamin D. The Vitamins, Am. Med. Assn., Chicago, Ill. P. 443.

Carver, J. S., Robertson, E. I., Brazie, D., Johnson, R. H., and St. John, J. L.: 1934. The vitamin D requirements of chickens. Wash. Agr. Exper. Sta., Bul. 299.

Correll, J. T., and Wise, E. C.: 1938. Studies on relative efficiency of vitamin D from several sources. II. Influence of vitamin D of different origins on the serum phosphatase of the chicken. Jour. Biol. Chem. 126:581.

Couch, J. R., Fraps, G. S., and Sherwood, R. M.: 1935. The vitamin D requirements of chickens grown in the absence of sunlight. Tex. Agr. Exper. Sta., Bul. 521.

——, Fraps, G. S., and Sherwood, R. M.: 1939. The vitamin D requirements for chicks. Fifty-second Ann. Rep. of the Tex. Agr. Exper. Sta., 113.

Doyle, L. P.: 1925. Rickets in mature chickens. Poultry Sci. 4:146.

——: 1940. Pathology of rickets in the fowl. Vet. Med. 35:248.

Drummond, J. C.: 1916. Observations upon the growth of young chickens under laboratory conditions. Biochem. Jour. 10:77.

Fell, H. B.: 1925. The histogenesis of cartilage and bone in the long bones of the embryonic fowl. Jour. Morph. and Physiol. 40:417.

Fritz, J. C., Archer, W., and Barker, D.: 1941. Vitamin D requirements of ducklings. Poultry Sci. 20:151.

Goodale, H. D.: 1926. Early growth rates of chickens with special reference to ultra-violet light. Am. Jour. Physiol. 79:44.

Hammond, J. C.: 1941. The vitamin D requirement of turkey poults. Poultry Sci. 20:204.

Hart, E. B., Halpin, J. G., and Steenbock, H.: 1922. The nutritional requirements of baby chicks. II. Further study of leg weakness in chickens. Jour. Biol. Chem. 52:379.

——, Steenbock, H., Lepkovsky, S., and Halpin, J. G.: 1923. The nutritional requirements of baby chicks. III. The relation of light to the growth of the chicken. Jour. Biol. Chem. 58:33.

——, Steenbock, H., Lepkovsky, S., Kletzien, S. W. F., Halpin, J. G., and Johnson, O. N.: 1925. The nutritional requirement of the chicken. V. The influence of ultra-violet light on the production, hatchability, and fertility of the egg. Jour. Biol. Chem. 65:579.

Hughes, J. S., and Payne, L. F.: 1924. Influence of ultra-violet light on young laying hens. Science 60:549.

——, Payne, L. F., and Latshaw, W. L.: 1925a. The influence of the ultra-violet light on leg weakness in growing chicks and on egg production. Poultry Sci. 4:151.

——, Payne, L. F., Titus, R. W., and Moore, J. M.: 1925b. The relation between the amount of ultra-violet light received by hens and the amount of antirachitic vitamin in the eggs produced. Jour. Biol. Chem. 66:595.

Innes, J. R. M., and Nicolaysen, R.: 1937. The assimilation of the Steenbock-Black diet in normal and vitamin D-deficient rats with and without caecum. Biochem. Jour. 31:101.

Jukes, T. H., and Sanford, T. D.: 1939. The vitamin D requirement of young turkeys. Jour. Nutr. 18:71.

Koch, E. M., and Koch, F. C.: 1941. The provitamin D of the covering tissues of chickens. Poultry Sci. 20:33.

Liu, S. H.: 1940. The role of vitamin D in the calcium metabolism in osteomalacia. Chinese Med. Jour. 57:101.

McGowan, J. P., and Emslie, A. R. G.: 1934. Rickets in chickens with special reference to its nature and pathogenesis. Biochem. Jour. 28:1503.

Maughan, G. H.: 1928. Ultra-violet wave lengths valuable in the cure of rickets in chickens. Am. Jour. Physiol. 87:381.

Murphy, R. R., Hunter, J. E., and Knandel, H. C.: 1936. The vitamin D requirements of growing chicks and laying hens. Pa. Agr. Exper. Sta., Bul. 334.

National Research Council: 1946. Recommended nutrient allowances for poultry. Mimeo. data No. 1.

Nicolaysen, R.: 1937a. Studies upon the mode of action of vitamin D. III. The influence of vitamin D on the absorption of calcium and phosphorus in the rat. Biochem. Jour. 31:122.

———: 1937b. Studies upon the mode of action of vitamin D. IV. The absorption of calcium chloride, xylose and sodium sulphate from isolated loops of the small intestine and of calcium chloride from the abdominal cavity in the rat. Biochem. Jour. 31:323.

———: 1937c. Studies upon the mode of action of vitamin D. V. The absorption of phosphates from isolated loops of the small intestine in the rat. Biochem. 31:1086.

——— and Jansen, J.: 1939. Vitamin D and bone formation in rats. Acta Paed. 23:405.

Nonidez, J. F.: 1928. Studies on the bones in avian rickets. I. Bone lesions in chickens deprived of the antirachitic factor after five weeks of normal growth. Am. Jour. Path. 4:463.

——— and Goodale, H. D.: 1927. Histological studies on the endocrines of chickens deprived of ultra-violet light. I. Parathyroids. Am. Jour. Anat. 38:319.

Pappenheimer, A. M., and Dunn, L. C.: 1925. The relation of leg weakness in growing chicks to mammalian rickets. Jour. Biol. Chem. 66:717.

Robertson, E. I., Rhian, M., and Wilhelm, L. A.: 1941. Growth and calcification of bronze turkey poults from hens fed different levels of vitamin D. Poultry Sci. 20:357.

Shipley, P. G.: 1924. The healing of rickety bones in vitro. Bul. Johns Hopkins Hosp. 35:304.

Sloan, H. J.: 1934. The seasonal variation in the antirachitic effectiveness of sunshine. Jour. Nutr. 8:731.

Smith, M. C., and Spector, H.: 1940. Further evidence of the mode of action of vitamin D. Jour. Nutr. 20:197.

Steenbock, H., Hart, E. B., Jones, J. H., and Black, A.: 1923. Fat-soluble vitamins. XIV. The inorganic phosphorus and calcium of the blood used as criteria in the demonstration of the existence of a specific antirachitic vitamin. Jour. Biol. Chem. 58:59.

Wilhelm, L. A., Robertson, E. I., and Rhian, M.: 1941. The effect of the level of vitamin D on egg production and hatchability of bronze turkey hens. Poultry Sci. 20:565.

## RIBOFLAVIN AND RIBOFLAVIN DEFICIENCY

Riboflavin was first known as vitamin $B_2$ and later as vitamin G. The chemical formula of this compound is very complex; the empirical formula is $C_{17}H_{20}N_4O_6$. It is a derivative of isoalloxazine, and as the name designates, one molecule of d-ribose is attached to the isoalloxazine (Booher, 1939). When in solution, it gives a yellow green fluorescent color. This color is readily detected in the egg white of hens that are receiving adequate amounts of riboflavin and is also noted in liquid whey. Riboflavin forms a phosphoric acid ester in the animal body; this in turn combines with a protein to form a yellow oxidation enzyme, which is present in all living cells of higher animal life, and is associated with chemical reactions which are involved in cell respiration. This vitamin is measured in micrograms of riboflavin and in chick units of vitamin G. These two units of measure are practically equal according to Norris and associates (1936). Jukes *et al.* (1938) state that a chick unit of vitamin G is equivalent to 2.3 micrograms of riboflavin.

SYMPTOMS AND PATHOLOGY OF RIBOFLAVIN DEFICIENCY

Symptoms of riboflavin deficiency in the chicks were first reported by Norris *et al.* (1930) and later by Bethke *et al.* (1931). These workers and also Lepkovsky and Jukes (1936) report the following riboflavin deficiency symptoms. When chicks are fed a diet deficient in riboflavin, they grow very slowly, become weak and emaciated; the appetite is fairly good, and diarrhea develops between the first and second week. The chicks do not walk, except when forced to do so and frequently walk upon their hocks with their toes curled inward (Fig. 7.8). When resting they usually sit on their hocks with the toes curled inward. If disturbed, they walk on their hocks with the aid of their wings. The wings often droop as though it were impossible for the chick to hold them in the normal position. The leg muscles are atrophied and flabby, and the skin
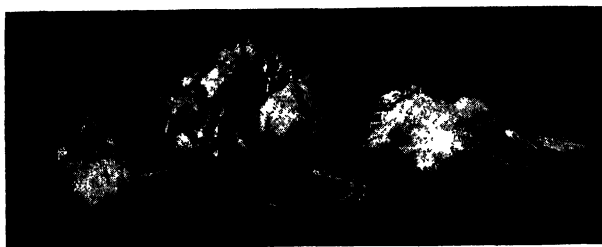


FIG. 7.8. Riboflavin deficiency (curly toe).

is dry and harsh. Young chicks in the advanced stages are unable to move around, and they lie with their legs sprawled out. Figure 7.9 shows a poult 35 days old, prostrate from riboflavin deficiency and with curled-toe paralysis. Figure 7.10 shows this same poult 5 days later after having been given two 100-microgram doses of riboflavin.

Post-mortem examination does not show any abnormalities of the internal organs, neither does bacteriological examination reveal any specific infection of the blood or other internal organs. In some cases, the thymus shows congestion and premature atrophy.

According to Davis *et al.* (1938a, 1938b) and Lepkovsky *et al.* (1938), the only symptoms noted of a deficiency of riboflavin in the diet of hens are decreased egg production, increased embryonic mortality, and an increase in the size and the fat content of the liver. The hatchability of eggs becomes very poor within two weeks after hens are fed a riboflavin deficient diet, but the hatchability improves to nearly normal within 7 days after adequate amounts of riboflavin are added to the diet. The embryos which fail to hatch from the eggs of hens on diets low in this factor are dwarfed, and show a high incidence of edema, degeneration of the Wolffian bodies, and a character-istically defective down. This type of down is referred to as "clubbed" and results from a failure of the down feathers to rupture the sheaths; this causes the feathers to coil and take the shape of a French knot.

Riboflavin deficiency in the young turkey is characterized by poor growth and incrustations in the corners of the mouth and on the eyelids. Severe

dermatitis of the feet and shanks, marked by edematous swelling, desquama-
tion, and deep fissures, appears in some of the deficient poults (Bethke and
Record, 1942). It is noted that these symptoms of riboflavin deficiency in
the turkey are similar to those of pantothenic acid deficiency in the chicken.

### HISTOPATHOLOGY OF RIBOFLAVIN DEFICIENCY

Phillips and Engel (1938) report that histologic examination of the
affected nerves shows definite degenerative changes in the myelin sheaths of
the main peripheral nerve trunks. This may be accompanied by axis cylinder



FIG. 7.9. A 35-day-old poult showing riboflavin deficiency with curled-toe paralysis.
(Richardson, Tex. Agr. Exper. Sta.)

swelling and fragmentation, Schwann cell proliferation, myelin changes,
gliosis, and chromatolysis in the spinal cord. In cases of curled-toe paralysis,
degeneration of the neuro-muscular end plate and muscle tissues are often
found. This indicates that riboflavin is necessary for the normal function of
the nervous system of the growing chick. Riboflavin is probably also essential
for myelin metabolism of the main peripheral nerve trunks (Phillips and
Engel, 1938). No gross dystrophy develops, although muscle fibers are in
some cases completely degenerated. The sciatic nerve is hypertrophied and
exhibits myelin degeneration in one or more of its branches. Similar changes
are apparent in some of the brachial nerve trunks.

In the case of embryos which fail to hatch from eggs laid by hens on riboflavin deficient diets, the nervous system shows degenerative changes very much like those described in riboflavin deficient chicks (Engel *et al.*, 1940).

### RIBOFLAVIN REQUIREMENTS OF CHICKENS

Workers in different laboratories are not in agreement as to the riboflavin requirements of chickens (Norris *et al.*, 1936; Heuser *et al.*, 1938; Lepkovsky and Jukes, 1935; Hunt *et al.*, 1939). The riboflavin or vitamin G requirements of chickens decrease with age and range from 1,580 micrograms of riboflavin or chick units of vitamin G per pound of feed for the second week, to 450 micrograms of riboflavin per pound of feed for the eighth week, according to Norris and co-workers (1936) and Heuser *et al.* (1938). Lepkovsky and Jukes (1935) reported that the chick required 450 chick units or 1,044 micrograms of riboflavin per pound of feed. Hunt *et al.* (1939) found that chicks required 860 to 910 micrograms of riboflavin per pound of feed for maximum growth to twelve weeks of age. Bethke and Record (1942) observed that it required 1,125 micrograms of riboflavin per pound of feed to give maximum growth to eight weeks but that it required 1,350 micrograms per pound of feed to prevent curled-toe paralysis. They also. found that naturally occurring and synthetic riboflavin were equally effective in promoting growth and preventing curled-toe paralysis. The riboflavin requirements may be higher than any of the figures given above for the prevention of histopathology of the nervous system resulting from riboflavin deficiency (Phillips and Engel, 1938). The riboflavin requirements for laying hens are higher when high hatchability is a factor than for egg production alone (Norris *et al.*, 1936; Davis *et al.*, 1938b).



FIG. 7.10. Same poult (Fig. 7.9) 5 days later after having received two 100-microgram doses of riboflavin. (Richardson, Tex. Agr. Exper. Sta.)

The Subcommittee on Poultry Nutrition of the National Research Council (1946) gives the requirements of riboflavin per pound of feed as follows:

for chicks up to eight weeks, 1,600 micrograms; eight to eighteen weeks, 900 micrograms; laying hens, 900 micrograms; and breeding hens, 1,300 micrograms of riboflavin per pound of feed.

### RIBOFLAVIN REQUIREMENTS OF DUCKS

The riboflavin requirements of ducklings is about 1,360 micrograms per pound of feed (Fritz *et al.*, 1939).

### RIBOFLAVIN REQUIREMENTS OF TURKEYS

The Subcommittee on Poultry Nutrition of the National Research Council (1946) gives the requirements of riboflavin for turkeys as follows: Poults up to eight weeks, 2,000 micrograms; eight to eighteen weeks, 900 micrograms; turkey hens, 900 micrograms; and turkey breeders, 1,600 micrograms of riboflavin per pound of feed.

### RIBOFLAVIN CONTENT OF FEEDS

The following table based on the work of Norris and associates (1936) gives the approximate number of micrograms of riboflavin per pound of feed with the exception of the value for cottonseed meal. The value for cottonseed meal was obtained from the work of Levine and Remington (1937) by using a conversion factor of three as suggested by Booher (1939) for the conversion of Bourquin-Sherman vitamin G units to micrograms of riboflavin.

In order to determine the riboflavin content of a diet, as illustrated in Table 3, one must multiply the number of pounds of each ingredient by the number of micrograms of riboflavin in a pound as given in Table 2.

This 114,660 micrograms of riboflavin per 100 pounds of feed would be satisfactory for chicks over eight weeks of age and for laying hens. It would not be satisfactory for baby chicks or for breeding hens. For baby chicks which seem to require 160,000 micrograms per pound of feed, the diet

TABLE 2

RELATIVE RIBOFLAVIN CONTENT OF COMMON FEEDSTUFFS USED IN POULTRY FEEDING

| Feedstuffs | Micrograms of Riboflavin per Pound |
|---|---|
| Dried yeast | 15,750 |
| Dried whey | 11,250 |
| Dried skimmilk | 7,650 |
| Alfalfa meal, dehydrated | 7,200 |
| Sardine fish meal | 3,150 |
| Meat scrap | 2,700 |
| Soybean oil meal | 1,350 |
| Wheat middlings, standard and flour | 900 |
| Wheat bran | 900 |
| Yellow corn | 450 |
| Cottonseed meal | 3,960 |

TABLE 3
Sample Diet

| Feed | Amount of Feed Used | Micrograms of Riboflavin per Pound of Feed | Total Micrograms of Riboflavin in Each Feed |
|---|---|---|---|
| Corn meal................ | 54 | 450 | 24,300 |
| Wheat gray shorts........ | 20 | 900 | 18,000 |
| Sardine meal............. | 6 | 3,150 | 18,900 |
| Cottonseed meal......... | 6 | 3,960 | 23,760 |
| Soybean oil meal........ | 6 | 1,350 | 8,100 |
| Alfalfa leaf meal......... | 3 | 7,200 | 21,600 |
| Minerals................. | 5 | ................ | ................ |
| Total............... | 100 | ................ | 114,660 |

studied would be short 45,340 micrograms (160,000 minus 114,660) of riboflavin per 100 pounds of feed. It would require about 4 pounds of dried whey instead of an equal amount of ground corn to supply this.

## REFERENCES ON RIBOFLAVIN

Bethke, R. M., Record, P. R., and Kennard, D. C.: 1931. A type of nutritional leg paralysis affecting chicks. Poultry Sci. 10:355.

—— and Record, P. R.: 1942. The relation of riboflavin to growth and curled-toe paralysis in chicks. Poultry Sci. 21:147.

Booher, L. E.: 1939. Chemical aspects of riboflavin. The Vitamins. Am. Med. Assn., Chicago, Ill. P. 249.

Boucher, R. V., Hill, F. W., Patrick, H., and Knandel, H. C.: 1941. The riboflavin requirement of turkeys for hatchability. Poultry Sci. 20:456.

Colorado Agricultural Experiment Station: 1938. Fifty-second Rep. P. 47.

Culton, T. G., and Bird, H. R.: 1940. The effect of some riboflavin supplements on chick growth and curled toe paralysis. Poultry Sci. 19:424.

Davis, H. J., Norris, L. C., and Heuser, G. F.: 1938a. The role of vitamin G in reproduction in poultry. Poultry Sci. 17:81.

——, Norris, L. C., and Heuser, G. F.: 1938b. Further evidence on the amount of vitamin G required for reproduction in poultry. Poultry Sci. 17:87.

Engel, R. W., Phillips, P. H., and Halpin, J. G.: 1940. The effect of a riboflavin deficiency in the hen upon embryonic development of the chick. Poultry Sci. 19:135.

Fritz, J. C., Archer, W., and Barker, D.: 1939. Riboflavin requirements of ducklings. Poultry Sci. 18:449.

Heuser, G. F., Wilgus, Jr., H. S., and Norris, L. C.: 1938. The quantitative vitamin G requirement of chicks. Poultry Sci. 17:105.

Hunt, C. H., Winter, A. R., and Bethke, R. M.: 1939. Further studies on the riboflavin requirements of the chicken. Poultry Sci. 18:330.

Jukes, T. H.: 1938. The vitamin G requirements of young poults. Poultry Sci. 17:227.

—— and Richardson, G. A.: 1938. Assays of riboflavin and the filtrate factor in certain milk products. Jour. Agr. Res. 57:603.

Lepkovsky, S., and Jukes, T. H.: 1935. The vitamin G requirements of the chick. Jour. Biol. Chem. 111:119.

—— and Jukes, T. H.: 1936. The response of rats, chicks, and turkey poults to crystalline vitamin G (flavin). Jour. Nutr. 12:515.

——, Taylor, L. W., Jukes, T. H., and Almquist, H. J.: 1938. The effect of riboflavin and the filtrate factor on egg production and hatchability. Hilgardia 11:559.

Levine, H., and Remington, R. E.: 1937. The vitamin G content of some foods. Jour. Nutr. 13:525.

National Research Council: 1946. Recommended nutrient allowances for poultry. Mimeo. data, No. 1.

Norris, L. C., Heuser, G. F., and Wilgus, Jr., H. S.: 1930. Is the chief value of milk for feeding poultry due to the presence of a new vitamin? Poultry Sci. 9:133.

——, Wilgus, Jr., H. S., Ringrose, A. T., Heiman, V., and Heuser, G. F.: 1936. The vitamin G requirement of poultry. Cornell Univ. Agr. Exper. Sta., Bul. 660.

Phillips, P. H., and Engel, R. W.: 1938. The histopathology of neuromalacia and "curled toe" paralysis in the chick fed low riboflavin diets. Jour. Nutr. 16:451.

Richardson, L. R.: 1947. Unpublished data. Tex. Agr. Exper. Sta.

## THIAMIN (B₁) AND THIAMIN DEFICIENCY

Thiamin (B₁) has an empirical formula of $C_{12}H_{18}N_4SOCl_2$ (Williams, 1939). It contains a pyrimidine nucleus, a thiazole group, and a primary alcohol group. This vitamin probably acts as a metabolite in carbohydrate metabolism and helps to remove and utilize pyruvic and lactic acids in this process.

The first deficiency disease to be produced experimentally was polyneuritis of the chicken in 1890 (Grijns, 1935). The fowl was used as the experimental animal in the early elucidation of beriberi. The avian species has been used as an experimental animal in numerous instances in the isolation and establishment of the structure of thiamin.

### THIAMIN DEFICIENCY IN FOWL FED A DIET OF POLISHED RICE

The published work on thiamin deficiency in poultry is somewhat confusing, because in most cases in the work prior to about 1930, a diet of polished rice was used to produce the deficiency disease (Grijns, 1935; Findlay, 1921; Vedder and Clark, 1912; McCarrison, 1918). Such a diet was later found to be deficient in factors other than the vitamin in question. It resulted in loss of appetite, extreme emaciation, and death. For that reason, research workers compared the symptoms of birds fed on polished rice with controls that were starved to death. It is, therefore, possible that the symptoms described for fowls fed a diet of polished rice may not all be true symptoms of thiamin deficiency.

Symptoms of polyneuritis or thiamin deficiency in the fowl appear 20 to 30 days after chickens are fed exclusively on polished rice (Findlay, 1921; Vedder and Clark, 1912). The onset of symptoms is somewhat gradual. There is a decided loss of weight in the affected bird accompanied by ruffled feathers, a blue comb, and leg weakness. Weakness of the legs is closely followed by an unsteady gait. As the deficiency continues, paralysis of the muscles develops, beginning with the flexors of the toes and progressing upward and affecting the extensor muscles of the legs, wings, neck, and the remainder of the body. The disease progresses from this stage with greater rapidity; the bird assumes a characteristic position in which it sits upon its flexed tarsometatarsus. If walking is attempted, it shuffles along upon the flexed tarsometatarsus; the extensor muscles are completely paralyzed. A general weakness develops with the paralysis, and the bird is usually no longer able to sit up but lies on its side. Many of the affected birds are unable to hold the wings in the accustomed position; the wing feathers drag on the ground. Retraction of the head is a frequent symptom as a result of paralysis of the anterior muscles in the neck.

The body temperature of the diseased birds is sometimes lowered from 105° to 109° F. in the normal fowl to 98° to 99° F., and in some cases even to

96° F. (Vedder and Clark, 1912). A progressive slowing of the respiratory rate and a hypertrophy of the adrenal glands, which appears to be more pronounced in the females than in the males, are common symptoms. The cortex is apparently affected to a greater extent than the medulla. Apparently the degree of hypertrophy in the adrenals determines the degree of edema or an increase of the water content of the tissues (McCarrison, 1918). In the chicken, edema occurs largely in the skin (Krause, 1922). It is also observed that the epinephrine content of the adrenals increases as this organ hypertrophies. A slight atrophy of the genital organs of birds affected with thiamin deficiency is somewhat more pronounced than in birds dying from starvation. Atrophy is more pronounced in the testes than in the ovaries. The heart shows a very slight degree of atrophy. The right side of the heart is frequently dilated; the auricle is more frequently affected than the ventricle. Considerable atrophy is noted of the muscular stomach and of the intestinal walls; this is so great in some cases that it may be visible to the naked eye. Extensive fatty degeneration of the liver is another lesion. Early workers in this field reported a degeneration of the nervous system (Kato and Shidzume, 1921; Gibson and Concepcion, 1914). Work by Engel and Phillips (1938), in which a diet complete in all factors except thiamin was fed, does not show any nerve degeneration in thiamin deficiency of the fowl. The nerve degenerations of the spinal cord and peripheral nerves reported in birds fed polished rice are traceable to the lack of vitamin A and riboflavin in the diet.

## REFERENCES ON THIAMIN

Engel, R. W., and Phillips, P. H.: 1938. The lack of nerve degeneration in uncomplicated vitamin B₁ deficiency in the chick and the rat. Jour. Nutr. 16:585.

Findlay, G. M.: 1921. An experimental study of avian beriberi. Jour. Path. and Bact. 24:175.

Gibson, R. B., and Concepcion, I.: 1914. A nerve degeneration in fowls fed on unhusked rice. Philippine Jour. Sci., Section B, 9:119.

Grijns, G.: 1935. Researches on Vitamins—1900–1911. Gorinchem-J. Noorduyn En Zoon N. V.

Kato and Shidzume: 1921. Pathological study on the nerves of the bird suffering from polished rice disease. Keio Med. Jour. [Abst. in Japan Med. World, 1:12 (1921) ].

Krause, D. J.: 1922. The water content of the tissues in experimental beriberi. Am. Jour. Physiol. 60:234.

McCarrison, R.: 1918. The pathogenesis of deficiency disease. Indian Jour. Med. Res. 6:275.

Vedder, E. B., and Clark, E.: 1912. A study of polyneuritis gallinarum. A fifth contribution to the etiology of beriberi. Philippine Jour. Sci., Section B, 7:423.

Williams, R. R.: 1939. The chemistry of thiamin (vitamin B₁). The Vitamins, Am. Med. Assn., Chicago, Ill. P. 141.

## PANTOTHENIC ACID AND PANTOTHENIC ACID DEFICIENCY

Pantothenic acid has been known by a number of different names; among these are the filtrate factor, as described by Lepkovsky and Jukes (1936) and Jukes and Lepkovsky (1936), the chick antidermatitis factor, named by Woolley et al. (1938), the third member of the vitamin B complex; and finally, upon the isolation, synthesis, and establishment of the structure of this compound, it was named pantothenic acid, by Williams and Major (1940). The deficiency caused by the lack of pantothenic acid was first re-

ported in 1930, but pantothenic acid was not synthesized and the structure established until 1940. The empirical formula is $C_9H_{17}NO_5$. It contains both a primary and a secondary alcohol group, a ketone group, and a carboxyl group.

### SYMPTOMS OF PANTOTHENIC ACID DEFICIENCY

Pantothenic acid deficient chicks are characterized by retarded and rough feather growth (Norris and Ringrose, 1930). The chicks are emaciated, and very definite crusty scab-like lesions appear in the corners of the mouth. The margins of the eyelids are granular, and small scabs develop on them. Williams and Major (1940) report that the eyelids are frequently stuck together by a viscous exudate; they are contracted, and vision is restricted (Fig. 7.11). In some cases the feathers are lost from the head. There is a slow sloughing of the keratinizing epithelium of the skin. The outer layers of skin between the toes and on the bottoms of the



FIG. 7.11. Pantothenic acid deficiency in 71-day-old chicks.

feet sometimes peel off, and small cracks and fissures appear at these points. These cracks and fissures enlarge and deepen, and the chicks move about very little. In some cases the skin layers of the feet of deficient chicks thicken and cornify, and wartlike protuberances develop on the balls of the feet.

Post-mortem examination shows the presence of a puslike substance in the mouth and an opaque grayish-white exudate in the proventriculus (Ringrose et al., 1931). The liver is hypertrophied and may vary in color from a faint yellow to a dirty yellow. The spleen is slightly atrophied. The kidneys are somewhat enlarged. The nerves and myelinated fibers of the spinal cord show myelin degeneration (Phillips and Engel, 1939). These degenerating fibers occur in all segments of the cord down to the lumbar region.

### REQUIREMENTS AND SOURCES OF PANTOTHENIC ACID

A definite relationship has been found between the pantothenic acid content of the diet and the pantothenic acid content of eggs and the concentra-

tion of this vitamin in the body tissues of chicks (Snell *et al.*, 1940, 1941).
An increase is noted in the pantothenic acid content of a hen's eggs within a
week after adding this vitamin to a pantothenic acid deficient diet. Lepkov-
sky *et al.* (1938) and Bauernfeind *et al.* (1939) report that a deficiency of
pantothenic acid in the diet does not appear to have any adverse effect on
the number of eggs laid, but it does result in a marked decrease in the hatch-
ability of the fertile eggs.

Growing chicks require between 2,700 micrograms of pantothenic acid
per pound of feed as recommended by Bauernfeind *et al.* (1942), and 6,500
micrograms per pound of feed as suggested by Jukes (1939). The Sub-
committee on Poultry Nutrition of the National Research Council (1946)
recommended 5,000 micrograms of pantothenic acid for growing chicks;
2,500 for laying hens, and 5,000 for breeding hens. Peanut meal, dried whey,
liver meal, yeast, dried milk, and rice bran are good sources of pantothenic
acid (Jukes and Lepkovsky, 1936; Jukes, 1937). Wheat middlings, corn, and
cottonseed meal are listed as poorer sources of pantothenic acid.

### REFERENCES ON PANTOTHENIC ACID

Bauernfeind, J. C., and Norris, L. C.: 1939. The role of the antidermatosis vitamin and a new water-soluble growth factor in the nutrition of the mature fowl. Jour. Nutr. 18:579.
——, Norris, L. C., and Heuser, G. F.: 1942. The pantothenic acid requirement of chicks. Poultry Sci. 21:142.
Jukes, T. H.: 1937. Further observations on the assay, distribution, and properties of the filtrate factor. Jour. Biol. Chem. 117:11.
——: 1939. The pantothenic acid requirement of the chick. Jour. Biol. Chem. 129:225.
—— and Lepkovsky, S.: 1936. The distribution of the "filtrate factor" (a water-soluble vitamin belonging to the vitamin B complex and preventing a dietary dermatitis in chicks) in certain feeding-stuffs. Jour. Biol. Chem. 114:117.
Kline, O. L., Keenan, J. A., Elvehjem. C. A., and Hart, E. B.: 1932. The use of the chick in vitamin $B_1$ and $B_2$ studies. Jour. Biol. Chem. 99:295.
Lepkovsky, S., and Jukes, T. H.: 1936. The effect of some reagents on the "filtrate factor" (a water-soluble vitamin belonging to the vitamin B complex and preventing a dietary derma-titis in chicks). Jour. Biol. Chem. 114:109.
——, Jukes, T. H., and Krause, M. E.: 1936. The multiple nature of the third factor of the vitamin B complex. Jour. Biol. Chem. 115:557.
——, Taylor, L. W., Jukes, T. H., and Almquist, H. J.: 1938. The effect of riboflavin and the filtrate factor on egg production and hatchability. Hilgardia 11:559.
National Research Council: 1946. Recommended nutrient allowances for poultry. Mimeo. data, No. 1.
Norris, L. C., and Ringrose, A. T.: 1930. The occurrence of a pellagrous-like syndrome in chicks. Science 71:643.
Phillips, P. H., and Engel, R. W.: 1939. Some histopathologic observations on chicks deficient in the chick antidermatitis factor or pantothenic acid. Jour. Nutr. 18:227.
Ringrose, A. T., Norris, L. C., and Heuser, G. F.: 1931. The occurrence of a pellagra-like syndrome in chicks. Poultry Sci. 10:166.
Snell, E. E., Pennington, D., and Williams, R. J.: 1940. The effect of diet on the pantothenic acid content of chick tissues. Jour. Biol. Chem. 133:559.
——, Aline, E., Couch, J. R., and Pearson, P. B.: 1941. The effect of diet on the pantothenic acid content of eggs. Jour. Nutr. 21:201.
Williams, R. J., and Major, R. T.: 1940. The structure of pantothenic acid. Science 91:246.
Woolley, D. W., Waisman, H. A., Mickelsen, O., and Elvehjem, C. A.: 1938. Some observations on the chick antidermatitis factor. Jour. Biol. Chem. 125:715.

### PYRIDOXINE (VITAMIN $B_6$) AND PYRIDOXINE DEFICIENCY

Pyridoxine (vitamin $B_6$) has been recognized since 1934 as the anti-
dermatitis factor of the rat but only recently has been shown to be neces-

sary for the chick (György, 1934; Carter and O'Brien, 1939; Hegsted *et al.*, 1939, 1940; Jukes, 1939). It is a pyridine with a rather simple empirical formula, $C_8H_{11}NO_3$ (Harris *et al.*, 1939).

### SYMPTOMS OF PYRIDOXINE DEFICIENCY IN THE CHICK

When chicks are fed a diet which is deficient in pyridoxine, the appetite is depressed, the food is not efficiently utilized, and the rate of growth is greatly decreased (Jukes, 1939; Hegsted *et al.*, 1940). The chicks have characteristic nervous symptoms, which consist of various convulsive movements. A jerky nervous movement of the legs results when the bird attempts to walk. In some cases, the symptoms of birds affected with this deficiency terminate in spasmodic convulsions and death. Pyridoxine has not been demonstrated to be necessary for egg production or for hatchability. Pyridoxine deficiency is not likely to occur frequently in the field, because the cereal grains are rich sources of this vitamin (Schneider *et al.*, 1939).

### REQUIREMENTS FOR PYRIDOXINE

Hogan *et al.* (1941) report that the requirements for White Leghorn chicks up to six weeks of age is 1,350 to 2,250 micrograms of pyridoxine per pound of feed. Briggs *et al.* (1942) estimate that only 1,250 to 1,350 micrograms of pyridoxine are required per pound of feed for White Leghorn chicks up to three weeks of age. Lucas *et al.* (1946), working with a cross between Rhode Island Red males and Barred Plymouth Rock females, found that 2,250 micrograms of pyridoxine per pound of feed was inadequate and suggested that possibly the requirements for this vitamin may vary with breeds and families. The Subcommittee on Poultry Nutrition of the National Research Council (1946) suggest that chickens require 1,600 micrograms of pyridoxine per pound of feed.

Boucher (1946) states that it appears unlikely that practical poultry rations composed of natural feed stuffs would be deficient in pyridoxine.

### REFERENCES ON PYRIDOXINE

Boucher, R. V.: 1946. Recent developments in turkey nutrition. Abst. Cornell Nutr. Conf., Cornell Univ., Ithaca. P. 54.

Briggs, Jr., G. M., Mills, R. C., Hegsted, D. M., Elvehjem, C. A., and Hart, E. B.: 1942. The vitamin $B_6$ requirement of the chick. Poultry Sci. 21:379.

Carter, C. W., and O'Brien, J. R.: 1939. Vitamin B complex in relation to the nutrition of the chick and pigeon. Proc. Seventh World's Poultry Cong. P. 126. ·

György, P.: 1934. Vitamin $B_2$ and the pellagra-like dermatitis in rats. Nature 133:498.

Harris, S. A., Stiller, E. T., and Folkers, K.: 1939. Structure of vitamin $B_6$. Jour. Am. Chem. Soc. 61:1242.

Hegsted, D. M., Oleson, J. J., Elvehjem, C. A., and Hart, E. B.: 1939. The "cartilage growth factor" and vitamin $B_6$ in the nutrition of chicks. Jour. Biol. Chem. 130:423.

———, Oleson, J. J., Elvehjem, C. A., and Hart, E. B.: 1940. The essential nature of a new growth factor and vitamin $B_6$ for chicks. Poultry Sci. 19:167.

Hogan, A. G., Richardson, L. R., Patrick, H.. O'Dell, B. L., and Kempster, H. L.: 1941. Vitamin $B_6$ and chick nutrition. Poultry Sci. 20:180.

Jukes, T. H.: 1939. Vitamin B₆ deficiency in chicks. Proc. Soc. Exper. Biol. and Med. 42:180.

Lucas, H. L., Heuser, G. F., and Norris, L. C.: 1946. The unexpected high requirements of chicks for pyridoxine. Poultry Sci. 25:137.

National Research Council: 1946. Recommended nutrient allowances for poultry. Mimeo. data, No. 1.

Schneider, H. A., Ascham, J. K., Platz, B. R., and Steenbock, H.: 1939. The anti-acrodynic properties of certain foods. Jour. Nutr. 18:99.

## ANTI-GIZZARD EROSION FACTOR

Gizzard erosion in chicks was first encountered in experiments dealing with vitamin K and at one time was thought to be due to a deficiency of this factor (Holst and Halbrook, 1933; Dam, 1934). The curative agent or compound for gizzard erosion has not been definitely isolated. A number of substances have been shown to cure the deficiency disease, but proof is lacking as to whether these compounds are the same as those found in feeds which have been shown to have curative properties for gizzard erosion (Bird *et al.*, 1939; Blount, 1939; Bird *et al.*, 1936; Almquist and Stokstad, 1936, 1937; Almquist and Mecchi, 1938; Almquist, 1938; Lansing and Miller, 1940; Crandall *et al.*, 1939; Esselen, 1939). Hatching conditions have been demonstrated to affect the occurrence and severity of gizzard erosion (Tepper and Bird, 1942).

### GROSS ALTERATIONS

Chicks affected with gizzard erosion show a degeneration of the horny layer of the gizzard (Almquist and Stokstad, 1936, 1937; Lansing *et al.*, 1939; Jungherr, 1935). This is usually preceded by hemorrhages varying in size from that of a pencil point to one many times this size. These may occur in any portion of the gizzard but are found more often in the cardiac end. In most cases, the eroded lining is loosened, usually brown or black, and presents a frayed appearance. The gizzard may be affected in one or more spots, and the size of these spots may vary from that of a pencil point to as large as a pencil. In some instances the lesions may be even larger (Fig. 7.12). Seventy to 96 per cent of the chicks from hens receiving good laying rations are affected with this abnormality when they hatch (Couch *et al.*, 1940; Charles *et al.*, 1941). It has also been found in the gizzards of embryos in late incubation stages. In embryos and in day-old chicks, the erosions are small, and in many cases, are apparent only as small blood clots or hemorrhages under the lining of the gizzard. As the chick grows older, the horny layer of the gizzard around these blood clots seems to break down, and the eroded areas enlarge; when the chicks are from five to eight weeks of age, the erosions usually disappear without adding any curative agent to the normal diet of the chicks. Since the condition appears in embryos and newly hatched chicks, it is evident that it is caused by a deficiency in the diet of the hen.

Jukes (1938) observed gizzard erosion in poults, but Esselen (1939) failed to produce this deficiency in young turkeys.

## HISTOPATHOLOGY

Histologic observations in the lining of the affected gizzard vary with the part of the gizzard from which sections are taken (Lansing *et al.*, 1939). In sections cut perpendicular to the attrition surface of the affected gizzard, a few polymorphonuclear cells and a large number of erythrocytes are observed in the secreted lining and also in the glands of affected areas. In a
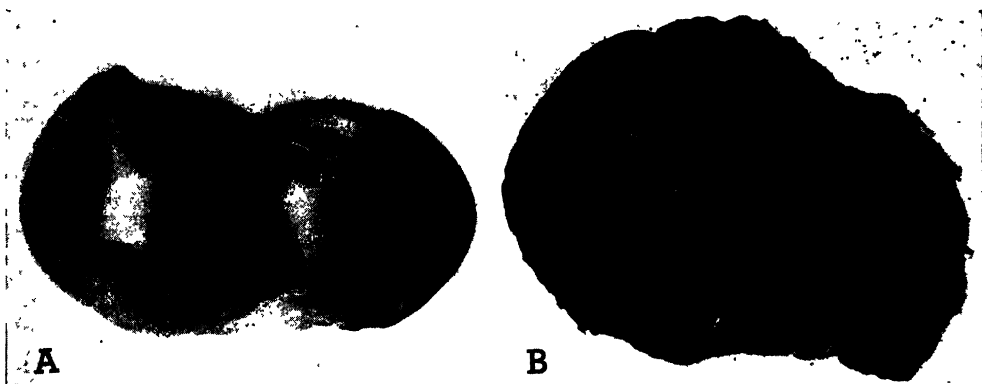


Fig. 7.12. A—Gizzard with a normal lining. B—Gizzard from a chick fed a diet high in corn. Note the roughened lining with the eroded areas. (Wilcke, Iowa State College.)

section taken through a slightly hemorrhagic area, blood cells are observed in the secretion from the glands. Blood cells occur in clumps; the glands apparently cease to function, and there is very little secretion above the glands or in the glandular cavity of areas severely affected with gizzard erosion. The glands are slightly shortened, curved, and twisted, and the tips in hemorrhagic areas are generally swollen. The epithelial cells are loosely arranged, and the epithelial lining of the glands shows indications of degeneration. Gizzard erosions are probably formed by the escape of blood into the secretion from which the lining is formed. The lining is weakened, and some of its cohesion is lost in these areas. The great muscular activity of the gizzard leads to the cracking and sloughing of the gizzard lining in the eroded spots. If the hemorrhage stops at this point, the gizzard lining is gradually rebuilt and assumes a normal appearance in about two weeks. If the original diapedesis is followed by pronounced hemorrhages, larger blood clots appear between the weakened lining and the glandular areas. This results in the formation of much larger eroded areas, and four weeks or more are required for the repair of these areas after the anti-gizzard erosion factor is incorporated in the diet.

## SOURCES OF THE ANTI-GIZZARD EROSION FACTOR

Wheat bran, fresh kale, oats, whole wheat, barley, and hempseed meal contain a factor which tends to prevent or cure gizzard erosion (Bird *et al.,* 1936; Almquist and Stokstad, 1936, 1937; Almquist, 1938; Esselen, 1939). In addition to the above, lesions of gizzard erosion have been cured by the feeding of cholic acid (Almquist and Mecchi, 1938; Bird *et al.,* 1938). One group of workers reported that chondroitin is an effective curative agent (Bird *et al.,* 1936, 1938); whereas another stated that chondroitin does not have any protective action (Crandall *et al.,* 1939). Others suggested that an alcohol extract of chicken feces cured gizzard erosion; the protective action in this case may be due to the bile acids recovered from the feces in the extraction process (Couch *et al.,* 1940).

Gizzard erosion has been produced in adult birds by ligation of the bile ducts (Lansing and Miller, 1940). Thus, the protective action of bile against gizzard erosion has been demonstrated in another manner.

### REFERENCES ON THE ANTI-GIZZARD EROSION FACTOR

Almquist, H. J.: 1938. The effect of hempseed preparations and of fineness of diet on the chick gizzard lining. Poultry Sci. 17:155.
——— and Mecchi, E.: 1938. The influence of bile on erosions of the chick gizzard lining. Jour. Biol. Chem. 126:407.
——— and Stokstad, E. L. R.: 1936. A nutritional deficiency causing gizzard erosions in chicks. Nature 137:581.
——— and Stokstad, E. L. R.: 1937. The gizzard factor of the chick. Jour. Nutr. 13:339.
Bird, H. R., Kline, O. L., Elvehjem, C. A., Hart, E. B., and Halpin, J. G.: 1936. The distribution and properties of the anti-gizzard-erosion factor required by chicks. Jour. Nutr. 12:571.
———, Oleson, J. J., Elvehjem, C. A., and Hart, E. B.: 1938. Effectiveness of chondroitin in preventing gizzard erosion in chicks. Jour. Biol. Chem. 126:671.
———, Oleson, J. J., Elvehjem, C. A., Hart, E. B., and Halpin, J. G.: 1939. Necessity for an organic dietary factor and for insoluble grit in the development of the gizzard lining in chicks. Proc. Seventh World's Poultry Cong. P. 174.
Blount, W. P.: 1939. Observations and experiments on gizzard erosion in poultry. Vet. Jour. 95:301.
Charles, T. B., Gillespie, J. H., Durgin, R. C., and Martin, C. I.: 1941. Incidence of gizzard erosion. Poultry Sci. 20:447.
Couch, J. R., James, L. E., and Sherwood, R. M.: 1940. Unpublished data. Tex. Agr. Exper. Sta.
Crandall, Jr., L. A., Chesley, F. F., Gray, R. E., and Robinson, H. E.: 1939. The effect of chondroitin sulfuric acid on gizzard erosion and growth in chicks. Jour. Nutr. 17:53.
Dam, H.: 1934. Hemorrhages in chicks reared on artificial diets: A new deficiency disease. Nature 133:909.
Esselen, Jr., W. B.: 1939. Nutritional gizzard lesions in chicks. Poultry Sci. 18:201.
Holst, W. F., and Halbrook, E. R.: 1933. A "scurvy-like" disease in chicks. Science 77:354.
Jukes, T. H.: 1938. The vitamin G requirements of young poults. Poultry Sci. 17:227.
Jungherr, E.: 1935. Diseases of brooder chicks. Storrs Agr. Exper. Sta., Bul. 202.
——— and Miller, D.: 1940. Dysfunction of the biliary system and hemorrhages in the gizzard of the chicken. Poultry Sci. 19:258.
Lansing, A. I., Miller, D., and Titus, H. W.: 1939. The formation of erosions of the gizzard lining in the young chick. Poultry Sci. 18:475.
Tepper, A. E., and Bird, H. R.: 1942. Gizzard lesions in day-old chicks. II. The time of origin and factors influencing the cause of gizzard lesions in chicks. Poultry Sci. 21:52.
Thompson, J. N., and Wilcke, H. L.: 1941. Gizzard erosion and feathering in chicks as influenced by the diet. Poultry Sci. 20:475.

## CHOLINE AND CHOLINE DEFICIENCY

Choline is a quaternary ammonium compound or nitrogenous base, which is a constituent of the phospholipids, lecithin, and sphingomyelin

(Bodansky, 1939). The empirical formula is $C_5H_{15}NO_2$. It has been suggested as a member of the vitamin $B_2$ complex (György and Goldblatt, 1940). In the mammal, choline has been shown to have a lipotropic action (Best and Ridout, 1939), to prevent a hemorrhagic degeneration of the kidney, and to function in the conversion of homocystine to methionine. When fowl are fed diets low in choline and high in fat, fatty infiltration of the liver does not occur (Kilborn, 1939).

### SYMPTOMS OF CHOLINE DEFICIENCY

When chicks or turkeys are fed diets deficient in choline, a condition known as perosis develops (Jukes, 1940a, 1940b; Jukes, 1941; Record and Bethke, 1941). Perosis is first characterized by pin-point hemorrhages and a slight puffiness about the hock joint. This is followed by an apparent flattening of the tibiometatarsal joint which is caused by a rotation or torsion of the metatarsus (Milne, 1936). The metatarsus continues to twist and may become bent or bowed so that it is out of alignment with the tibia. When this condition exists, the leg cannot adequately support the weight of the bird. The articular cartilage is displaced, and the tendons, principally the gastrocnemius, which it encloses slips from the condyles. Choline may also be considered as necessary for normal bone metabolism. In addition to the above, the blood and bone phosphatase of chicks affected with perosis are depressed below that of normal ones (Wiese *et al.*, 1939). According to Abbott and DeMasters (1940), when laying pullets are fed diets deficient in choline, increased mortality, increased abortion of egg yolks from the ovaries, and an increase in the percentage of fatty acids in the liver result. The percentage of fatty acids in the livers of choline deficient birds is much higher in females than in males.

### REQUIREMENTS FOR CHOLINE

Melass *et al.* (1946) fed choline in amounts from .05 per cent to 4 per cent to chicks from one to ten weeks of age. Increasing amounts of choline reduced the subcutaneous, abdominal, and mesenteric fat deposits in the chick. Their work shows that it is doubtful if normal, practical rations for chicks are deficient in choline.

The Subcommittee on Poultry Nutrition of the National Research Council (1946) recommends that the choline requirements for chicks up to eight weeks of age is 700 milligrams per pound of feed and for poults of this age it is 900 milligrams of choline.

Lucas *et al.* (1946) point out that very good production and satisfactory hatchability have been observed with practical breeding rations containing as little as .12 per cent or 550 milligrams of choline per pound of feed. They also point out that the requirement for choline is effected by the amino acid

content of the ration and by the fact that birds are able to synthesize choline in the intestinal tract.

More work is needed to determine whether practical rations composed of a variety of feeds are deficient in choline.

<div align="center">REFERENCES ON CHOLINE</div>

Abbott, O. D., and DeMasters, C. U.: 1940. Choline in the diet of chickens. Jour. Nutr. 19:47.
Best, C. H., and Ridout, J. H.: 1939. Choline as a dietary factor. Ann. Rev. Biochem. 8:349.
Bodansky, M.: 1939. Introduction to Physiological Chemistry. John Wiley and Sons, Inc., New York. Pp. 74, 207.
Branion, H. D.: 1938. Minerals in poultry nutrition. Scient. Agr. 18:217.
du Vigneaud, V., Chandler, J. P., Moyer, A. W., and Keppel, D. M.: 1939. The effect of choline on the ability of homocystine to replace methionine in the diet. Jour. Biol. Chem. 131:57.
Griffith, W. H., and Wade, N. J.: 1939. Some effects of low choline diets. Proc. Soc. Exper. Biol. and Med. 41:188.
György, P., and Goldblatt, H.: 1940. Choline as a member of the vitamin B₂ complex. Jour. Exper. Med. 72:1.
Jukes, T. H.: 1940a. Prevention of perosis by choline. Jour. Biol. Chem. 134:789.
————: 1940b. Effect of choline and other supplements on perosis. Jour. Nutr. 20:455.
————: 1941. Studies of perosis in turkeys. I. Experiments related to choline. Poultry Sci. 20:251.
Kilborn, L. G.: 1939. Choline and liver fat in birds. Chinese Jour. Physiol. 14:283.
Lucas, H. L., Norris, L. C., and Heuser, G. F.: 1946. Observations on the choline requirements of hens. Poultry Sci. 25:373.
Melass, V. H., Pearson, P. B., and Sherwood, R. M.: 1946. Toxicity of choline in the diet of growing chicks. Proc. Soc. Exper. Biol. and Med. 62:174.
Milne, H. I.: 1936. Studies of perosis in chicks. Proc. Sixth World's Poultry Cong. 2:155.
National Research Council: 1946. Recommended nutrient allowances for poultry. Mimeo. data, No. 1.
Record, P. R., and Bethke, R. M.: 1941. Preliminary observations on choline in chick nutrition. Poultry Sci. 20:471.
Sherwood, R. M., and Couch, J. R.: 1933. Feeding for efficient growth and prevention of slipped tendons in chickens. Tex. Agr. Exper. Sta., Bul. 476.
Wiese, A. C., Johnson, B. C., Elvehjem, C. A., Hart, E. B., and Halpin, J. G.: 1939. A study of blood and bone phosphatase in chick perosis. Jour. Biol. Chem. 127:411.

<div align="center">VITAMIN E AND VITAMIN E DEFICIENCY</div>

<div align="center">ENCEPHALOMALACIA AND MYOPATHY</div>

Vitamin E deficiency was first demonstrated in 1922 with rats (Todd, 1939; Smith, 1940). In 1930 it was observed in chicks fed a purified diet. In 1935 and 1936 vitamin E deficiency was found in chicks under field conditions (Jungherr, 1936). The condition has been encountered experimentally in a number of laboratories, both in this country and abroad (Pappenheimer and Goettsch, 1931; Wolf and Pappenheimer, 1931; Hogan and Shrewsbury, 1930; Pappenheimer et al., 1939; Ni, 1937; Hogan and Boucher, 1933; Elvehjem et al., 1937) ; it may be of wider occurrence in the field than some workers realize. It has been demonstrated that there is a factor in cod-liver oil which hinders the utilization of vitamin E by the chick (Hammond, 1941) ; this may cause the occurrence of encephalomalacia in chicks, under field conditions, that seem to be receiving a satisfactory diet and that do not appear to be affected otherwise.

Pappenheimer and Goettsch (1934a, b) , Seifried and Heidegger (1936) , and Pappenheimer et al. (1939) report that the same experimental diet

which has caused vitamin E deficiency (encephalomalacia) in chicks causes myopathy in ducklings and myopathy of the gizzard of turkeys.

### ENCEPHALOMALACIA OF CHICKENS

#### SYMPTOMS

According to Pappenheimer and Goettsch (1931), Hogan and Shrewsbury (1930), Ni (1937), Adamstone (1934, 1936), Dam and Glavind (1939), and Bird and Culton (1940), in experimentally produced cases the chicks first appear droopy, their eyes are closed, and the birds assume a fixed posture for long periods. This is followed by a nervous derangement, which is often intensified by excitement, or it may occur very suddenly without apparent cause. The features most often observed are ataxia or lack of power to coordinate muscular movements, retraction of the head, sometimes with lateral twisting, forced movements, increasing incoordination, a rapid contraction and relaxation of the legs, and finally complete prostration and death (Fig. 7.13).



FIG. 7.13. Encephalomalacia.

Even under these conditions, complete paralysis of the wings or legs is not observed. The deficiency usually manifests itself between the fifteenth and the thirtieth day of the chick's life, although it has been known to occur as early as the seventh day and as late as the fifty-sixth day. No difference was noted between the susceptibility of the various breeds studied (Pappenheimer *et al.*, 1939; Pappenheimer and Goettsch, 1933). Male and female chicks are equally susceptible.

Vitamin E apparently is not necessary for egg production in the fowl (Holmes and Cravens, 1940a, b), but is necessary for embryonic development (Adamstone, 1931; Adamstone and Card, 1934). Embryos from hens fed diets in which vitamin E has been destroyed die on or before the fourth day of incubation. The development of the embryos is retarded, and there is a marked underdevelopment of the hematopoietic system. A lethal ring formed by the rapid growth of the mesoderm hems in or chokes off the vitelline, arteries, and veins, and causes the death of the embryo.

The male fowl is not affected by vitamin E deficiency as is the male rat (Adamstone and Card, 1934). Cockerels kept on a diet deficient in vitamin E

for as long as two years have still been able to fertilize eggs; however, testicular degeneration and even complete atrophy of the testis may occur.

The pathological lesions consist of encephalomalacia (Pappenheimer and Goettsch, 1931; Pappenheimer *et al.*, 1939). The cerebellum, the cerebral hemispheres, the medulla, and the midbrain are most commonly affected in the order named. In chicks which are killed soon after the appearance of symptoms of encephalomalacia, the cerebellum is softened, swollen, and the meninges are edematous. Minute hemorrhages are often visible on the surface of the cerebellum. The convolutions are flattened. In some cases, as much as four-fifths of the cerebellum may be affected, while in others lesions may be so small that they cannot be recognized grossly. A day or two after the symptoms of encephalomalacia are first manifested, the necrotic areas present a greenish-yellow opaque appearance. Healing sometimes occurs spontaneously, in which case the affected areas are shrunken and depressed below the surface of the healthy tissue, and the color changes to brownish-yellow.

In the cerebrum, the necrotic tissue is frequently pale, swollen, and wet, and in the early stages becomes sharply delineated from the remaining normal tissue (Pappenheimer *et al.*, 1939). Some cases are so affected that the greater portion of both hemispheres in the cerebrum are destroyed. Other cases are so mildly affected that the lesions are apparent only on microscopic examination. In the cerebrum, the affected tissue is greenish-yellow, but when healing has occurred, the color changes to a rusty brown.

Medullary lesions are not so readily noted in a macroscopic examination (Pappenheimer *et al.*, 1939). A flattening and general swelling of the ventral surface indicates the presence of internal lesions. It is estimated that after one is familiar with the disease a macroscopic diagnosis can be correctly made in approximately 90 per cent of the cases.

According to Wolf and Pappenheimer (1931) and Pappenheimer *et al.* (1939), the microscopic lesions of the cerebellum are very characteristic but are variable in extent and distribution. In all cases, edema in the beginning is followed by capillary hemorrhages, thrombosis, and necrosis of neuroglial elements and ganglion cells. There is an increase in weight and moisture content of the part or parts of the brain affected. Edema is probably the most constant and striking feature; it results in a squaring of the convolutions and obliteration of the sulci. Edema is apparent in the Purkinje cell zone and results in a cribriform or vacuolar structure. The Purkinje cells, Golgi cells, and small cells of the granular layer undergo a degeneration known as

ischemic necrosis. The Purkinje cells become angular and narrow and lose their Nissl substances; their nuclei become pyknotic. Nuclei of the Bergmann cells are hydropic and swollen.

One of the earliest recognizable lesions of encephalomalacia is a circulatory disturbance in the brain (Wolf and Pappenheimer, 1931; Pappenheimer et al., 1939). The pial vessels and the capillaries of the granular and molecular layers and the capillaries of the central white matter in the region of the cerebellum are engorged with erythrocytes. Not all vessels are affected in this manner; some vessels are distended while others are collapsed. This is particularly true in the white matter but less often in the granular layers, and is only occasionally noted in the lower portion of the molecular layer. Large numbers of small hemorrhages are found in the pia and cerebellar layers and are very conspicuous in the lower portion of the molecular layer. The fibers of the white matter are separated by a mild edema. Hyaline capillary thrombi appear even in the most recent and small areas of necrosis and are a very constant feature of the lesions. When the capillaries undergo repair, the endothelial cells become greatly swollen and grow very actively; they quite often sprout laterally into the necrotic tissues. The lesions in the cerebrum, midbrain, and medulla are very much like those of the cerebellum. Cerebral lesions appear to result from vascular disturbances, but the vascular disturbances are not associated with significant alterations in cell plasma ratio, plasma, or blood volume (Pappenheimer et al., 1939; Pappenheimer and Graff, 1932).

### REQUIREMENTS AND RECOMMENDATIONS

The requirements of vitamin E have not been sufficiently worked out to make definite recommendations, and as suggested earlier, these recommendations will probably be influenced by the amount of cod-liver oil in the ration, since it was demonstrated that there is a factor in cod-liver oil that hinders the utilization of vitamin E by chickens (Hammond, 1941).

It has been demonstrated that in addition to alpha-tocopherol, encephalomalacia is largely prevented by a number of vegetable oils (Pappenheimer et al., 1939; Ni, 1937; Goettsch and Pappenheimer, 1936; Pappenheimer and Goettsch, 1934a, b; Dam et al., 1938; Babcock and Jukes, 1937). Among these are cottonseed oil, peanut oil, wheat germ oil, soybean oil, and the nonsaponifiable fraction of soybean oil. These oils have been demonstrated to contain alpha-tocopherol.

### NUTRITIONAL MYOPATHY IN DUCKLINGS

Ducklings are affected differently from chicks (Pappenheimer et al., 1939; Pappenheimer and Goettsch, 1934a, b). In ducks there is a universal degeneration of the skeletal muscles. Other organs and tissues do not appear

to be involved. The deficiency manifests itself in the second and third weeks and is characterized by the birds walking awkwardly with the feet turned in. They are found sprawled flat on the floor of their pens. Affected ducklings placed on their backs have difficulty in assuming the normal upright position. They finally become so weak that they are not able to raise their heads. The eyes are somewhat sunken, and in occasional cases there are coarse tremors and athetoid movements. The head is never retracted; there are no forced movements, and no spasticity.

### GROSS PATHOLOGY

The skeletal muscles are very pale in color in contrast to the normal dark red (Pappenheimer and Goettsch, 1934a; Pappenheimer et al., 1939). The muscle tissue is watery and translucent, and the contractility is impaired. Microscopic examination shows a varying proportion of necrotic fibers. These exhibit the usual appearance of hyaline or waxy degeneration. Even in the severe cases there is practically no cellular change. The intermuscular tissue is edematous. The creatine content of the muscles of affected ducklings is reduced. The moisture content is increased, which is in accord with the edema noted.

### REQUIREMENTS AND RECOMMENDATIONS

Nutritional myopathy of ducks is prevented by feeding alpha-tocopherol, hydrogenated cottonseed oil, wheat germ oil, soybean oil, and possibly other vegetable oils containing alpha-tocopherol (Pappenheimer, 1940; Pappenheimer et al., 1939). Some work has already been done on the requirements, but more is needed before definite recommendations can be given.

### NUTRITIONAL MYOPATHY OF THE GIZZARD IN TURKEYS

Characteristic lesions are located in the smooth muscle of the gizzard, and they appear as circumscribed gray areas, which are of firmer texture than the normal muscle, and in some cases, suggest scar tissue (Jungherr and Pappenheimer, 1937; Pappenheimer et al., 1939). In most cases, these lesions are recognizable grossly. Often irregular, grayish patches are also observed through the transparent serous covering of the gizzard even though the external conformation and tonus of the gizzard appear normal.

### HISTOPATHOLOGY

Histologic examination shows hyaline degeneration; the muscle fibers are swollen, lumpy, fragmented, and separated by edema (Pappenheimer et al., 1939; Jungherr and Pappenheimer, 1937). The nuclei appear shrunken and pyknotic and eventually disappear completely throughout the necrotic area. There is fibrous replacement of the muscle fibers characterized

by very abrupt changes of the bundles and septa. Muscle fibers in areas which recently suffered pathological changes frequently cannot be distinguished from one another. A secondary inflammatory reaction is occasionally found in and around some of the necrotic areas. The inflammatory cells in these infected areas are mostly pseudoeosinophils. These tend to accumulate in the intermuscular septa and perivascular connective tissue, and are scattered among the necrotic fibers.

### REQUIREMENTS AND RECOMMENDATIONS

Nutritional myopathy of turkeys appears to be controlled by the same carriers of alpha-tocopherol that were effective with chicks, namely, cottonseed oil, wheat germ oil, soybean oil, and probably others (Pappenheimer *et al.,* 1939) .

### REFERENCES ON VITAMIN E

Adamstone, F. B.: 1931. The effects of vitamin E deficiency on the development of the chick. Jour. Morph. and Physiol. 52:47.

———: 1934. The effects of severe and prolonged vitamin E deficiency in the chick. Anat. Record 60, Am. Soc. Zool. Proc. 36.

———: 1936. A brain disorder in young chicks fed on a diet treated with ferric choloride for the destruction of vitamin E. Anat. Record 67, Am. Soc. Zool. Proc. 106.

——— and Card, L. E.: 1934. The effects of vitamin E deficiency on the testis of the male fowl (*Gallus domesticus*) . Jour. Morph. 56:339.

Babcock, Jr., S. H., and Jukes, T. H.: 1937. Beneficial effect of nonsaponifiable fraction of soybean oil on chicks fed a simplified diet. Proc. Soc. Exper. Biol. and Med. 36:720.

Bird, H. R., and Culton, T. G.: 1940. Generalized edema in chicks prevented by d, l-alpha-tocopherol. Proc. Soc. Exper. Biol. and Med. 44:543.

Dam, H., and Glavind, J.: 1939. Alimentary exudative diathesis and its relation to vitamin E. Skand. Arch. Physiol. 82: 299.

———, Glavind, J., Bernth, O., and Hagens, E.: 1938. Anti-encephalomalacia activity of dl-alpha-tocopherol. Nature 142:1157.

Elvehjem, C. A., Phillips, P. H., and Hart, E. B.: 1937. Differentiation between $B_1$ deficiency and "encephalomalacia" in growing chicks. Proc. Soc. Exper. Biol. and Med. 36:129.

Goettsch, M., and Pappenheimer, A. M.: 1936. The prevention of nutritional encephalomalacia in chicks by vegetable oils and their fractions. Jour. Biol. Chem. 114:673.

Hammond, J. C.: 1941. A factor in cod liver oil that hinders the utilization of vitamin E by chickens. Poultry Sci. 20:369.

Hogan, A. G., and Boucher, R. V.: 1933. The nutritional requirements of the chick. Mo. Agr. Exper. Sta., Res. Bul. 198.

——— and Shrewsbury, C. L.: 1930. Deficiencies of synthetic diets in chick nutrition. Jour. Nutr. 3:39.

Holmes, C. E., and Cravens, W. W.: 1940a. The effect of feeding wheat germ oil. I. Egg production and hatchability. Poultry Sci. 19:303.

——— and Cravens, W. W.: 1940b. The effect of feeding wheat germ oil. II. Growth, age to sexual maturity, and egg production. Poultry Sci. 19:311.

Jungherr, E.: 1936. A field condition resembling nutritional encephalomalacia in chicks. Science 84:559.

——— and Pappenheimer, A. M.: 1937. Nutritional myopathy of the gizzard in turkeys. Proc. Soc. Exper. Biol. and Med. 37:520.

Ni, T. G.: 1937. The prevention of nutritional encephalomalacia by gelatin. Chinese Jour. Physiol. 12:281.

———: 1938. Further experiments on the prevention of nutritional encephalomalacia in chickens. Chinese Jour. Physiol. 13:229.

Pappenheimer, A. M.: 1940. Prevention of nutritional myopathy of ducklings by alphatocopherol. Proc. Soc. Exper. Biol. and Med. 45:457.

——— and Goettsch, M.: 1931. A cerebellar disorder in chicks, apparently of nutritional origin. Jour. Exper. Med. 53:11.

——— and Goettsch, M.: 1933. Nutritional encephalomalacia in chicks. Influence of age, growth, and breed upon susceptibility. Jour. Exper. Med. 57:365.

——— and Goettsch, M.: 1934a. Nutritional myopathy in ducklings. Jour. Exper. Med. 59:35.

Pappenheimer, A. M., and Goettsch, M.: 1934b. Protection afforded by certain vegetable oils against nutritional encephalomalacia of chicks. Preliminary report. Proc. Soc. Exper. Biol. and Med. 31:777.

———, Goettsch, M., and Jungherr, E.: 1939. Nutritional encephalomalacia in chicks and certain related disorders of domestic birds. Storrs Agr. Exper. Sta., Bul. 229.

——— and Graff, S.: 1932. Blood volume in normal chicks and in chicks with nutritional encephalomalacia. Proc. Soc. Exper. Biol. and Med. 30:321.

Seifried, O., and Heidegger, E.: 1936. Untersuchungen über eine enzootisch auftretende Muskeldystrophie bei jungen Enten. Arch. f. wiss. und prakt. Tiereilk. 70:122.

Smith, L. I.: 1940. The chemistry of vitamin E. Chem. Rev. 27:287.

Todd, A. R.: 1939. The chemistry of vitamin E. A symposium held under the auspices of the Food Group (Nutrition Panel) of the Society of Chemical Industry. Keppl Street, London, W.C. 1, England. P. 3.

Wolf, A., and Pappenheimer, A. M.: 1931. The histopathology of nutritional encephalomalacia of chicks. Jour. Exper. Med. 54:399.

## VITAMIN K AND VITAMIN K DEFICIENCY

Vitamin K deficiency in chicks was first observed in Denmark (Dam, 1929). The work of Almquist (1939) shows that this vitamin is also required by chicks, ducks, geese, and pigeons, but that it is not important in practical poultry feeding, because it is abundant in poultry feeds. Although the chick was used as the experimental animal in a large amount of the vitamin K research, the findings have already been applied to some extent in human medicine (Andrus and Lord, 1940; Clark et al., 1939). Two chemical forms of vitamin K have been isolated; both of them are napthoquinones. The side chains are different in the two forms. A large number of compounds of structure similar to vitamin K have been shown to exhibit vitamin K activity; some of these are much more active than vitamin K as originally isolated (Doisy et al., 1940; Fieser, 1940; Ansbacher et al., 1940; Almquist and Klose, 1940).

### SYMPTOMS OF VITAMIN K DEFICIENCY

Symptoms of vitamin K deficiency appear most frequently in from 15 to 20 days after the chicks are placed on a diet free of vitamin K (Clark et al., 1939; McFarlane et al., 1931a, b; Almquist and Stokstad, 1935; Dam and Schönheyder, 1934). Hemorrhages appear on the breast, legs, and wings. Intramuscular hemorrhages also occur on the legs, and in some cases large hemorrhages are found in the abdominal cavity. Anemia, which follows the hemorrhages, is a result of the loss of blood and is not due to a reduction in the hemoglobin content of the blood. The blood of chicks affected with vitamin K deficiency fails to clot on standing; this is due to a decreased prothrombin content. Within 4 to 6 hours after vitamin K or a vitamin K concentrate is fed to deficient chicks, the blood clots normally.

### SOURCES OF VITAMIN K

The best sources of vitamin K are green leafy plants, dehydrated alfalfa leaf meal, or cereal grasses, carrot tops, and soybean oil; the amount of vitamin K is greatly reduced in dried alfalfa exposed to sunlight (Almquist,

1939; Cravens *et al.,* 1941) . A direct relationship exists between the amount of vitamin K in the diet of the hen and the amount in the yolks of her eggs and the amount stored in the newly hatched chicks (Almquist, 1939; Almquist and Stokstad, 1936) . It has been difficult to produce vitamin K deficiency in mammals, because the vitamin is synthesized by bacteria in the digestive tract. It has been observed that the amount of vitamin K synthesis in the intestinal tract and the symptoms of vitamin K deficiency demonstrated in the mammal are influenced by the type of intestinal flora present (Dam and Glavind, 1939) .

Vitamin K is found in the feces of chicks even though they are fed a diet free of vitamin K; the amount in the feces increases within the first 24 hours after the droppings are voided. This indicates that the vitamin was synthesized to some extent in the lower portion of the intestinal tract of the chicks and more extensively in the feces of the chicks for the first 24 hours after being voided.

### REFERENCES ON VITAMIN K

Almquist, H. J.: 1939. Properties of vitamin K. Proc. Seventh World's Poultry Cong. P. 138.
———— and Klose, A. A.: 1940. Comparative activities of certain anti hemorrhagic compounds. Proc. Soc. Exper. Biol. and Med. 45:55.
———— and Stokstad, E. L. R.: 1935. Dietary hemorrhagic disease in chicks. Nature 136:31.
———— and Stokstad, E. L. R.: 1936. Factors influencing the incidence of dietary hemorrhagic disease in chicks. Jour. Nutr. 12:329.
Andrus, W. DeW., and Lord, Jr., J. W.: 1940. Correction of prothrombin deficiencies by means of 2-methyl-1, 4-napthoquinone injected intramuscularly. Jour. Am. Med. Assn. 114:1336.
Ansbacher, S., Fernholz, E., and Dolliver, M. A.: 1940. Vitamin K-active derivatives of 2-methyl-1, 4-napthohydroquinone. Jour. Am. Chem. Soc. 62:155.
Clark, Jr., R. L., Dixon, C. F., Butt, H. R., and Snell, A. M.: 1939. Deficiency of prothrombin associated with various intestinal disorders: Its treatment with the antihemorrhagic vitamin (vitamin K) . Proc. Staff Meet. of the Mayo Clinic 14:407.
Cravens, W. W., Randle, S. B., Elvehjem, C. A., and Halpin, J. G.: 1941. Vitamin K studies. I. Effect of the vitamin K content of the hen's ration on the clotting ability of chick blood. Poultry Sci. 20:313.
Dam, H.: 1929. Cholesterinstoffwechsel in Hühnereiern und Hühnchen. Biochem. Zeitschr. 215:475.
———— and Glavind, J.: 1939. Alimentary K-avitaminosis in rats. Zeitschr. Vitaminforschg. 9:71.
———— and Schönheyder, F.: 1934. A deficiency disease in chicks resembling scurvy. Biochem. Jour. 28:1355.
Doisy, E. A., Binkley, S. B., Thayer, S. A., and McKee, R. W.: 1940. Vitamin K. Science 91:58.
Fieser, L. F.: 1940. The synthesis of vitamin K₁. Science 91:31.
McFarlane, W. D., Graham, Jr., W. R., and Hall, G. E.: 1931. Studies in protein nutrition of the chick. I. The influence of different protein concentrates on the growth of baby chicks, when fed as the source of protein in various simplified diets. Jour. Nutr. 4:331.
————, Graham, Jr., W. R., and Richardson, F.: 1931. The fat soluble vitamin requirements of the chick. I. The vitamin A and D vitamin content of fish meal. Biochem. Jour. 25:358.

### ASCORBIC ACID (VITAMIN C) AND ASCORBIC ACID DEFICIENCY

Ascorbic acid is not required in the diets of chickens, ducks, geese, turkeys, guinea fowls, pheasants, or pigeons (Hart *et al.,* 1925; Carrick and Hauge, 1925; Plimmer *et al.,* 1923; and Hauge and Carrick, 1926) . Chickens fed diets deficient in this vitamin show no ill effects, and their eggs hatch normally. The egg is practically devoid of ascorbic acid, yet the chick embryo contains large quantities of this vitamin as early as the fourth day of incubation (Ray, 1934) . The liver and kidneys of chickens fed a diet low

in ascorbic acid for long periods of time contain large quantities of this factor (Hart *et al.*, 1925; Carrick and Hauge, 1925; Plimmer *et al.*, 1923). It seems evident that ascorbic acid is necessary for the normal metabolic processes in birds, and that birds are able to synthesize quantities of the vitamin sufficient for the normal body processes (Ray, 1934).

## REFERENCES ON ASCORBIC ACID

Carrick, C. W., and Hauge, S. M.: 1925. Presence of the antiscorbutic substance in the livers of chickens fed on scorbutic diets. Jour. Biol. Chem. 63:115.

Hart, E. B., Steenbock, H., Lepkovsky, S., and Halpin, J. G.: 1925. The nutritional requirement of the chicken. VI. Does the chicken require vitamin C? Jour. Biol. Chem. 66:813.

Hauge, S. M., and Carrick, C. W.: 1926. The antiscorbutic vitamin in poultry nutrition. Poultry Sci. 5:166.

Plimmer, R. H. A., Rosedale, J. L., and Raymond, W. H.: 1923. The rearing of chickens on the intensive system. Part IV. C-vitamin requirements of chickens and other birds. Biochem. Jour. 17:787.

Ray, S. N.: 1934. A note on the presence of vitamin C in the chick embryo. Biochem. Jour. 28:189.

## BIOTIN AND BIOTIN DEFICIENCY

Biotin deficiency was first produced in the chick by Lease and Parsons (1934). This was not recognized until the reports of Hegsted *et al.* (1940, 1942) were published. During the latter period, a number of workers, including Eakin *et al.* (1940), McElroy and Jukes (1940), Ansbacher and Landy (1941), Jukes and Bird (1942), and Richardson *et al.* (1942), discovered the importance of biotin in chick nutrition. Cravens *et al.* (1942, 1944) found that biotin was essential for embryonic development of the hen's egg and made a careful study of the effect of biotin deficiency on embroyonic development. Patrick *et al.* (1941) reported that biotin was necessary for the prevention of dermatitis in turkey poults.

### SYMPTOMS OF BIOTIN DEFICIENCY IN THE CHICK

The symptoms of biotin deficiency in the chick have been described by Hegsted *et al.* (1940). There are two types of dermatitis lesions: those which appear on the feet, and those which appear around the beak and eyes. When chicks are fed a biotin deficient diet, lesions appear in about three weeks, the growth curve flattens out, and the bottoms of the feet become rough and calloused and may be severely affected before mandibular lesions are evident. As the syndrome progresses the entire bottom of the foot becomes encrusted, and hemorrhagic cracks appear together with skin fissures. The toes may become necrotic and slough; the top of the foot and leg are not so severely affected and may exhibit only a dry scaliness. Lesions appear in the vicinity of the mouth somewhat later than the first signs of the deficiency on the feet. The mandibular lesions may spread to include the entire area around the beak, which usually has rather large encrustations in the corners of the mouth and which are more severe than those noted in pantothenic acid avitaminosis.

Perosis is also a characteristic deficiency symptom of biotin avitaminosis. This has been demonstrated by a number of workers including McElroy and Jukes (1940), Jukes and Bird (1942), and Richardson *et al.* (1942). Symptoms of perosis are described elsewhere in this text.

## BIOTIN DEFICIENCY IN HENS

Cravens *et al.* (1942, 1944) reported that hens could be maintained on biotin deficient diets without deficiency symptoms appearing in the birds. Egg production apparently was not affected. However there was a decided effect on hatchability with one peak of embryonic mortality occuring during the first week, and a second peak of embryonic mortality occuring during the last 3 days of the incubation period. Characteristic embryonic abnormalities were observed. Embryos from hens fed biotin deficient diets developed syndactyly, an extensive webbing between the third and fourth toes. These workers also observed that a large number of the embryos which failed to hatch were chondrodystrophic, and were characterized by a reduced size, a parrot beak, severely crooked tibia, and/or a much shortened or twisted tarsometatarsus.

## BIOTIN DEFICIENCY IN POULTS

Symptoms of biotin deficiency in young poults were noted and described by Patrick *et al.* (1941). These symptoms are very similar to those described above for chicks insofar as the dermatitis of the feet and mandibles are concerned. These workers did not refer to perosis in their report.

## REQUIREMENTS FOR BIOTIN

Hegsted *et al.* (1942) reported that the chick required at least 32–45 micrograms of biotin per pound of ration. It would probably be safer to recommend 90 micrograms of biotin per pound. From the work of Cravens *et al.* (1944) 90 micrograms of biotin per pound of ration would also be sufficient to meet the requirements of laying hens.

The poultry subcommittee of the National Research Council (1946) recommends 45 micrograms of biotin per pound of feed for starting feeds for chicks up to eight weeks of age and 70 micrograms for breeding hens.

## REFERENCES ON BIOTIN

Ansbacher, S., and Landy, M.: 1941. Biotin and scaly dermatosis of the chick. Proc. Soc. Exper. Biol. and Med. 48:3.

Eakin, E. E., McKinley, W. A., and Williams, R. J.: 1940. Egg-white injury in chicks and its relationship to a deficiency of vitamin H (Biotin). Science 92:224.

Cravens, W. W., McGibbon, W. H., and Sebesta, E. E.: 1944. Effect of biotin deficiency on embryonic development in the domestic fowl. Anat. Record 90:55.

————, Sebesta, E. E., Halpin, J. G., and Hart, E. B.: 1942. Effect of biotin on reproduction in the domestic fowl. Proc. Soc. Exper. Biol. and Med. 50:101.

Hegsted, D. M., Oleson, J. J., Mills, R. C., Elvehjem, C. A., and Hart, E. B.: 1940. Studies on a
    dermatitis in chicks distinct from pantothenic acid deficiency. Jour. Nutr. 20:599.
———, Mills, R. C., Briggs, G. M., Elvehjem, C. A., and Hart, E. B.: 1942. Biotin in chick
    nutrition. Jour. Nutr. 23:175.
Jukes, T. H., and Bird, F. H.: 1942. Prevention of perosis by biotin. Proc. Soc. Exper. Biol.
    and Med. 49:231.
Lease, J. G., and Parsons, H. T.: 1934. The relationship of dermatitis in chicks to a lack of
    Vitamin B, and egg-white. Biochem. Jour. 28:2109.
National Research Council: 1946. Recommended nutrient allowances for poultry. Mimeo. data,
    No. 1.
McElroy, L. W., and Jukes, T. H.: 1940. The formation of the anti-egg-white injury factor
    (Biotin) in the rumen of the cow. Proc. Soc. Exper. Biol. and Med. 45:296.
Patrick, H., Boucher, R. V., Dutcher, R. A., and Knandel, H. C.: 1941. Biotin and prevention of
    dermatitis in turkey poults. Proc. Soc. Exper. Biol. and Med. 48:456.
Richardson, L. R., Hogan, A. G., and Miller, O. N.: 1942. The relation of biotin to perosis in
    chicks. Mo. Agr. Exper. Sta., Res. Bul. 343.

### NICOTINIC ACID AND NICOTINIC ACID DEFICIENCY

Elvehjem *et al.* (1938) discovered that nicotinic acid was a vitamin, and since then it has been shown that many animals require it in their diet. Ringrose *et al.* (1938) reported that poults require some factor other than the known ones to prevent perosis. Briggs *et al.* (1942) showed that chicks fed a highly purified diet require nicotinic acid. Briggs (1946) demonstrated that the turkey requires nicotinic acid in its diet. Evans *et al.* (1943) reported that poults require a factor or factors other than choline to completely prevent perosis.

#### SYMPTOMS OF NICOTINIC ACID DEFICIENCY

Briggs (1946) describes the symptoms of nicotinic acid deficiency as inflammation of the mouth, diarrhea, low feed consumption, low feed utilization, slow growth, poor feathering, and perosis. Briggs *et al.* (1946) showed that tryptophane supplements the action of nicotinic acid. The work of Rosen *et al.* (1946) and that of Singal *et al.* (1946) indicate that in rats tryptophane may be a precursor to nicotinic acid synthesis.

#### REQUIREMENTS FOR NICOTINIC ACID

The requirements of nicotinic acid in a diet is difficult to estimate; the amount required varies with the tryptophane content of the ration. It is suggested by Briggs *et al.* (1946) that White Leghorn chicks require about 8 milligrams of nicotinic acid per pound of feed. Briggs *et al.* (1946) state that New Hampshire chicks may require about 23 milligrams of nicotinic acid per pound of diet. This would appear to be the upper limit needed, since this figure was arrived at by using diets high in gelatin and other materials which were selected specifically to be low in tryptophane. The Subcommittee on Poultry Nutrition of the National Research Council (1946) recommends 8 milligrams of nicotinic acid per pound of feed for starting chickens up to eight weeks of age.

Briggs (1946) suggests that poults require from 13 to 23 milligrams of nicotinic acid per pound of feed to prevent perosis and other deficiency

symptoms. Higher levels may be necessary for optimum growth. Boucher (1946) estimates that the nicotinic acid requirement of poults is 23 milligrams per pound of feed. Boucher (1946) also reports "It appears unlikely that a practical ration composed of natural feed stuffs would be deficient in nicotinic acid." He also states that commercial feeds are protected against nicotinic acid deficiency by such feeds as skimmed milk, buttermilk, whey, liver meal, glandular meals, brewer's dried yeast, fermentation solubles, and fish solubles.

## REFERENCES

Boucher, R. V.: 1946. Recent developments in turkey nutrition. Abst. Cornell Nutr. Conf., Cornell Univ., Ithaca, N. Y.

Briggs, Jr., G. M.: 1946. Nicotinic acid deficiency in turkey poults and the occurrence of perosis. Jour. Nutr. 31:79.

————, Groschke, A. C., and Lillie, R. J.: 1946. Effects of proteins low in tryptophane on growth of chickens and on laying hens receiving nicotinic acid-low rations. Jour. Nutr. 32:659.

————, Mills, R. C., Elvehjem, C. A., and Hart, E. B.: 1942. Nicotinic acid in chick nutrition. Proc. Soc. Exper. Biol. and Med. 51:59.

Elvehjem, C. A., Madden, R. J., Strong, F. M., and Woolley, D. W.: 1938. The isolation and identification of the anti-black tongue factor. Jour. Biol. Chem. 123:137.

Evans, R. J., Rhian, J. M., and Draper, C. I.: 1943. Perosis in turkey poults and the choline content of their diets. Poultry Sci. 22:88.

National Research Council: 1946. Recommended nutrient allowances for poultry. Mimeo. data, No. 1.

Ringrose, A. T., Martin, J. H., and Insko, Jr., W. M.: 1939. Manganese requirements of turkey poults. Poultry Sci. 18:409.

Rosen, F., Huff, J. W., and Perlzweig, W. A.: 1946. The effect of tryptophane on the synthesis of nicotinic acid in the rat. Jour. Biol. Chem. 163:343.

Singal, S. A., Briggs, A. P., Sydensricker, V. P., and Littlejohn, J.: 1946. Effect of tryptophane on urinary excretion of nicotinic acid in rats. Fed. Proc. 5:154.

## FOLIC ACID AND FOLIC ACID DEFICIENCY

Folic acid, a member of the B-complex, is one of the newer vitamins to be recognized, isolated, and synthesized. It includes both pteroylglutamic acid and pteroyltriglutamic acid (Jukes and Stokstad, 1947). Stokstad and Manning (1938) were among the first workers to find that this vitamin was necessary for chicks. They called it "Factor U." A factor in liver which prevented anemia in chicks was described by Hogan and Parrott (1940). They called this vitamin $B_c$.

The following year Mitchell et al. (1941) reported a factor which they called folic acid that prevented anemia in chicks; a concentrate of folic acid was produced by them from spinach. Hutchings et al. (1944) found that anemia in chicks was prevented by the L. casei factor.

Day et al. (1945) reported that their monkey vitamin M and the L. casei factor are both folic acid conjungates. Charkey (1945) reported that the Cornell R factor was one or more folic acid conjugates. Recent work suggests that all of these factors are either folic acid or one or more of the acid conjugates.

The study of folic acid has been more difficult than some other vitamins, because appreciable quantities of this vitamin are synthesized in the intestinal

tract of animals; the amount synthesized is affected by both feed and medication or drugs (Daniel *et al.*, 1946).

### SYMPTOMS OF FOLIC ACID DEFICIENCY

Richardson *et al.* (1945) reported that a deficiency of folic acid caused poults to develop a type of cervical paralysis which usually resulted in death. When first noted the neck is extended and rigid. Symptoms are shown in Figures 7.14 and 7.15. First the attacks are intermittant, later they are continuous. The wings quiver and droop slightly, and the poult chirps as if in pain. The bird may have diarrhea with thin, white excreta. Death results within 1 or 2 days after the severe symptoms appear.



Fig. 7.14. Folic acid deficiency, showing "straight neck" paralysis. (Richardson and Hogan, Mo. Agr. Exper. Sta.)

Richardson *et al.* (1945) reported further that, of ten poults fed a folic acid deficient diet, five showed typical cervical paralysis and died within a day or two after the severe symptoms appeared. Four others died with no characteristic symptoms and one of the ten lived to fourteen weeks of age. In later experiments with larger numbers of poults the results were very similar. The poults of Richardson *et al.* (1945) showed a lower red-cell volume than is normal, but none were extremely anemic.

Deficiency of folic acid in turkeys was also reported by Jukes (1947) to result in slow growth and a moderate degree of anemia.

### REQUIREMENTS FOR FOLIC ACID

Robertson *et al.* (1946) reported that, for chicks, the total folic acid requirement for survival up to six weeks of age is about 115 micrograms per pound of feed; for hemoglobin formation and growth up to four weeks of

age the requirement is about 200 micrograms; for hemoglobin formation to six weeks only about 160 micrograms per pound of feed are required, but for feather growth at six weeks of age, 250 micrograms of folic acid per pound of feed are required. Russell and Taylor (1947) state that poults require up to 910 micrograms of folic acid per pound of feed for normal feathering, hemoglobin, and for the prevention of cervical paralysis.

Little and Briggs (1947) suggest that New Hampshire chicks receiving synthetic diets require from 775 to 900 micrograms of folic acid per pound of synthetic diet. Robertson (1946) reports that limited data on commercial starting rations show that they contain more than 450 micrograms of folic acid per pound of ration, which indicates that they should not be deficient



FIG. 7.15. Folic acid deficiency, showing "straight neck" paralysis. (Richardson and Hogan, Mo. Agr. Exper. Sta.)

in folic acid. Jukes and Stokstad (1947) report that New Hampshire chicks were found to utilize pteroylglutamic acid or pteroyltriglutamic acid (a conjugated derivative which occurs in feeds) equally well for growth and prevention of anemia. Boucher (1946) states that it is unlikely that practical poultry rations composed of natural feedstuffs would be deficient in folic acid.

Work by Schweigert et al. (1948) indicates that while the turkey hens receiving .2 milligrams of folic acid per pound of feed seemed to show no deficiency symptoms, and that they layed well and that the eggs hatched satisfactorily, the poults showed higher mortality and slower growth than when the mother turkeys received 1.0 milligram per pound of feed.

## REFERENCES

Boucher, R. V.: 1946. Recent developments in turkey nutrition. Abst. Cornell Nutr. Conf., Cornell Univ., Ithaca, N. Y.

Charkey, L. W.: 1945. Factor R and its relation to the other members of the B complex. Doctoral thesis. Cornell Univ.

Daniel, L. J., Farmer, F. A., and Norris, L. C.: 1946. Folic acid and perosis. Jour. Biol. Chem. 163:349.

Day, P. L., Mims, V., and Trotter, J. R.: 1945. The relationship between vitamin M and other *Lactobacillus casei* factors. Jour. Biol. Chem. 161:45.

Hogan, A. G., and Parrott, E. M.: 1940. Anemia in chicks caused by a vitamin deficiency. Jour. Biol. Chem. 132:507.

Hutchings, B. L., Stokstad, E. L. R., Bohonos, N., and Slobodkin, N. H.: 1944. Isolation of a new *Lactobacillus casei* factor. Science 99:371.

Jukes, T. H., and Stokstad, E. L. R.: 1947. The comparative utilization of pteroylglutamic acid and pteroyltriglutamic acid by chicks on purified diets. Jour. Biol. Chem. 168:563.

———, Stokstad, E. L. R., and Belt, M.: 1947. Deficiencies of certain vitamins as studied with turkey poults on a purified diet. Jour. Nutr. 33:1.

Lillie, R. J., and Briggs, G. M.: 1947. Folic acid requirements of New Hampshire chicks receiving synthetic diets. Poultry Sci. 26:295.

Mitchell, H. K., Snell, E. E., and Williams, R. J.: 1941. Concentration of "folic acid." Jour. Am. Chem. Soc. 63:2284.

Richardson, L. R., Hogan, A. G., and Kempster, H. L.: 1945. Requirement of the turkey poult for vitamin $B^1_c$. Jour. Nutr. 30:151.

Robertson, E. I., Daniel, L. J., Farmer, F. A., Norris, L. C., and Heuser, G. F.: 1946. The folic acid requirements of chicks for growth, feathering, and hemoglobin formation. Proc. Soc. Exper. Biol. and Med. 62:97.

Russell, W. C., and Taylor, M. W.: 1947. Folic acid requirements of turkey poults on a purified ration. Proc. Biol. Chem. Div. Am. Chem. Soc., Atlantic City, N. J. P. 41b.

Schweigert, B. S., German, H. L., Pearson, P. B., and Sherwood, R. M.: 1948. The effect of the pteroylglutamic acid intake on the performance of turkeys and chickens. Jour. Nutr. 25:89.

Stokstad, E. L. R., and Manning, P. D. V.: 1938. Evidence of a new growth factor required by chicks. Jour. Biol. Chem. 125:687.

# PULLORUM DISEASE

By HENRY VAN ROEKEL, *Department of Veterinary Science, Massachusetts Agricultural Experiment Station, Amherst, Massachusetts*

☆  ☆  ☆

In 1899 the etiological agent of pullorum disease was discovered by Rettger (1900). He first described the disease as a "Fatal Septicemia of Young Chicks," but later (1909) he designated it as "White Diarrhea." However, in that same year in a subsequent report by Rettger and Stoneburn (1909) they applied the term "Bacillary White Diarrhea" in order to distinguish it from other avian diseases which might be classified under a common terminology of "White Diarrhea" as was reported by Jones (1911).

In 1929 at the Second Annual Meeting of Investigators and Workers in Bacillary White Diarrhea Control, Rettger (1932), at the suggestion received from a research member of the Pennsylvania Department of Agriculture, proposed that the term "Pullorum Disease" be substituted for "Bacillary White Diarrhea." This new terminology was internationally adopted because of its brevity, specificity, and appropriateness in designating a disease entity which affected not only chicks but also mature poultry and fowl other than chickens.

## HISTORICAL

The isolation of the causative agent of pullorum disease by Rettger (1900) in 1899 gave an insight into the serious chick-raising problem reported in the lay-press (E. A. H., 1905; Gifford, 1905; Graham, 1904) which appeared before and after the epoch-making discovery. At the close of the nineteenth century, this malady was considered a very serious menace to the poultry industry. During the first decade of the twentieth century, investigators definitely proved that pullorum disease was an egg-borne infection. The cycle of infection involved an infected hen laying infective eggs, hatching infected chicks, which could develop into mature infected stock.

During the second decade, Jones (1913b), and later others (Gage, Paige, and Hyland, 1914; Rettger, Kirkpatrick, and Jones, 1914), announced the practical application of the macroscopic tube agglutination test for the detection of "carriers" of the organism. The application of this test in the control and eradication of the disease was carried out rather extensively in some of

the eastern states so that toward the close of the second decade, official state testing programs were inaugurated.

The progressive development and expansion of the baby chick industry through more modern methods of incubation, brooding, and transportation have influenced the dissemination of the disease. Incubator transmission has been and still is an important factor in the spread of pullorum disease from plant to plant by means of flock replacements.

Another event which has influenced the pullorum status throughout the world was the organization of the Conference of Investigators and Workers in Bacillary White Diarrhea Control (W. R. H., 1928), composed first of representatives from the New England States and later (Anon., 1930) enlarged to include other eastern states and provinces in Canada. This Conference has made a concerted effort to bring about standardization and uniformity of methods and to stimulate an interest in the practical eradication of the disease from breeding flocks.

The Conference of Research Workers in Animal Diseases of North America (Anon., 1933) formulated "Standard Methods of Diagnosis of Pullorum Disease in Barnyard Fowl" which were adopted by that organization and also by the United States Livestock Sanitary Association in 1932. These methods of diagnosis have served as valuable guides in the combat against pullorum disease.

Schaffer, MacDonald, Hall, and Bunyea (1931) announced the development of the modified whole-blood method in which stained antigen is employed. In view of its apparent simplicity, it has been widely used with the result that many infected birds have been detected, and thus their removal from breeding flocks was made possible.

The United States Department of Agriculture inaugurated the National Poultry Improvement Plan which represents a national effort to control and eradicate pullorum disease in poultry flocks (Anon., 1941).

The above-mentioned events point out that during the past fifty years the poultry industry has been made more secure through the publication of these scientific facts and through the adoption of official Federal and State plans which are being employed to combat pullorum disease.

## DISTRIBUTION AND ECONOMIC IMPORTANCE

Pullorum infection is world-wide in its distribution. It is likely to be found wherever poultry is being raised. Prior to the date the organism was discovered by Rettger (1900), the disease had been observed in the United States and Canada by those engaged in poultry husbandry. Later reports (Reis and Nobrega, 1936) revealed the recognition of the disease in England, different parts of continental Europe, South Africa, South America, Australia, and Japan.

Economic losses of serious proportions were reported during the last decade in the nineteenth century. As the poultry industry developed, especially the breeding and hatching phases, the incidence of the disease was permitted to become greater, and the infection was disseminated more widely throughout the United States and Canada. During a later period this likewise was true in other countries.

Losses from the disease may be experienced through severe chick mortality, reduced fertility and hatchability, retardation in growth, reduced egg production, increased mortality among adult stock, and a reduction in the sales quality of the stock.

Rettger (1900, 1901) in his early observations of the disease found that among hen-reared and among artificially brooded chicks, total mortalities might approximate 85 per cent during the first four weeks of age. Later reports (Jones, 1911; Kaupp, 1917) reveal similar or even higher losses among chicks. Attempts to raise chicks from breeding stock affected with pullorum disease usually meet with failure and disappointment. Losses among different hatches of chicks from the same breeding stock may vary considerably. In some cases, little or no mortality may be encountered among chicks raised from infected breeding stock. However, many extrinsic factors may be associated with the infecting organism in producing the disease in chicks.

Pullorum disease definitely causes a reduction in fertility and hatchability. Bushnell *et al.* (1926), Dearstyne *et al.* (1929), and Runnells (1929) reported that more infertile eggs are laid by infected than by noninfected birds. However, some infected birds may lay a normal number of fertile eggs which hatch well. Beaudette *et al.* (1923), Bushnell *et al.* (1926), Dearstyne *et al.* (1929), and Runnells (1929) observed that hatchability might be very seriously affected in eggs laid by infected birds. In one instance, a difference of 18.2 per cent in the hatchability of fertile eggs laid by reacting and nonreacting birds was reported in favor of the latter. This aspect of the disease is very significant from the economic point of view since large commercial hatcheries purchase hatching eggs at a premium over market price of eggs used for food. In some instances, bonuses based on the fertility and hatchability of the eggs are paid to the producer. In order to obtain maximum profit from breeding birds, it is important that they be free of infection.

All chicks which have suffered from an outbreak of pullorum infection do not necessarily succumb nor do all necessarily make complete recovery. Some chicks are definitely stunted in their growth, and in a pullorum infected chick flock, all the same age, they may exhibit a great variation in size and growth rate. Van Roekel (1931) reported that among twenty-nine six-weeks-old chicks exposed to infection when 72 hours old, the range in weight varied from 90 to 558 grams. In some instances, it is advisable to destroy the

survivors, clean and disinfect the quarters, and replace with clean stock. Chicks retarded in their growth do not mature into vigorous well-developed laying or breeding birds.

Occasionally, mature stock may suffer extensive mortalities from pullorum infection. Jones (1913a) reported an acute outbreak among adult fowl with a mortality of approximately 8 per cent during one month. Infertile eggs and dead embryos received from an infected flock, which were fed to the adult flock, were regarded as the source of infection. Plastridge and Rettger (1930) described a pleomorphic type of *S. pullorum* which was highly virulent and produced an acute septicemia in mature fowls, marked pathologic changes, and a mortality of from 5 to 25 per cent. This outbreak occurred in a flock known to be free of the disease for seven consecutive years. Acute outbreaks have resulted in some flocks in which the infection was regarded as dormant among the carriers in the flock. Chronic carriers of the infection appear to be less able to withstand changes in environmental conditions and concurrent flock diseases, with the result that a higher mortality is found among carriers of the disease.

Losses also result from a decrease in egg production from infected adults, according to Runnells (1929), Doyle (1925), Dearstyne *et al.* (1929), and Asmundson and Biely (1930). The latter report that the average first-year egg production for nonreacting and reacting birds was 221 and 160 eggs, respectively. A greater variation in production was noted among reactors than among the nonreactors. The egg production for the reactors was significantly lower than that of the nonreactors in every one of the twelve months. On the contrary, Dearstyne *et al.* (1929) found that the reactor in its first-year egg production may be profitable and that variations in production may be expected depending upon the localization and degree of infection. It is generally accepted that pullorum infection may impair production and should for that reason, as well as for other reasons, not be tolerated in a flock.

Through the inauguration of official control programs and the establishment of official grades for pullorum-tested flocks, it has been made possible to identify flocks as to their official pullorum disease status and thereby afford the buying public an opportunity to buy stock of the highest quality. In certain areas, stock from pullorum disease-free flocks commands a higher price and is in greater demand than stock from flocks which have revealed infection.

### ETIOLOGY

Pullorum disease is a bacterial infection caused by an organism which Rettger (1900, 1909) first designated as a bacillus and a few years later named *Bacterium pullorum.*

More recently the systematic bacteriologist has classified the etiological

agent in the "Salmonella" genus, and at the present time, the organism is recognized as *Salmonella pullorum*. It has many features in common with other members of the Salmonella group.

The organism is a long, slender rod (.3–.5 × 1–2.5μ) with slightly rounded ends (Fig. 8.1–3). It readily stains with ordinary basic anilin dyes and is Gram negative. The cells occur singly, with chains of more than two bacilli being rarely found. An occasional filament and large cell may be found in smear prepara-
tions. It is nonmotile, non-
liquefying, nonchromo-
genic, nonsporogenic, and
facultatively anaerobic.
Optimum growth occurs at
37° C. and under normal
atmospheric conditions.
On meat extract agar (pH
7.0–7.2) heavily seeded
with inoculum, the colo-
nies appear discrete,
smooth, glistening, ho-
mogeneous, entire, dome-
shaped, transparent, and
varying in form from
round to angular (Fig.
8.1–1). On chicken infu-
sion agar, the growth is
slightly more luxuriant,
with colonies possessing a
lesser degree of transpar-



Fig. 8.1. 1—Isolated colony on primary culture. Meat extract agar. 40 hours old. ×15. 2—Isolated colony on primary culture. Liver infusion agar. 40 hours old. ×15. 3—Cells in a smear prepared from colony illustrated in 1. ×1,200. 4—Cells in a smear prepared from colony illustrated·in 2. ×1,200.

ency. On liver infusion agar, the growth is even more luxuriant and markedly translucent (Fig. 8.1–2 and 4). Congested colonies remain small (1 mm. or less), but isolated colonies may attain a diameter of 3 to 4 mm. or more. Surface markings may appear as the colony increases in size and age, but as a rule the young colony on a heavily seeded plate changes little with age.

The following substances are attacked with acid and with or without gas production: arabinose, dextrose, galactose, levulose, mannite, mannose, rhamnose, and xylose. Substances not attacked include adonite, dextrin, dulcitol, erythrol, glycerol, inositol, inulin, lactose, raffinose, saccharose, salicin, sorbite, and starch. Maltose is attacked very infrequently as has been reported by Edwards (1928), Hendrickson (1927), Hinshaw (1941), and Pacheco and Rodrigues (1936). However, the results in some instances were

attributed to the materials and methods employed for the cultivation of the organism. Edwards (1928) concluded that acid production in maltose by *S. pullorum* was made possible through the hydrolization of the sugar by the alkali that slowly developed upon prolonged incubation. Hendrickson (1927) observed that when serum water was used for sugar base, maltose was fermented by *S. pullorum*. Pacheco and Rodrigues (1936) encountered similar findings and claimed the acid production by the organism was the result of serum-enzyme hydrolysis of the sugar. Van Roekel (1935) reported a laboratory strain which had been considered as a maltose nonfermenting organism, but after a lapse of several years since its original isolation it acquired and retained the ability to attack maltose. No plausible explanation could be given for the sudden change in the maltose-fermenting characteristic. Subsequent investigations (Van Roekel *et al.*, 1937) revealed that strains which possessed a potential tendency to ferment maltose could be identified by cultivating them in a maltose-peptone solution for a period of time. Strains undergoing a change in behavior toward maltose would exhibit red and white colonies when plated on a modified Endo's medium (maltose substituted for lactose). Strains that produced both maltose-fermenting and nonmaltose-fermenting colonies exhibited only nonmaltose-fermenting colonies after being subjected to animal passage. An apparently pure maltose-fermenting strain did not lose this property when subjected to animal passage. It is apparent that *S. pullorum* may display variation in its behavior in the fermentation of maltose, and for that reason this sugar cannot be regarded of value in the identification of the organism. Variation in the behavior of some strains may be observed occasionally, especially in regard to gas production. Litmus milk remains practically unchanged. Indol and acetyl-methyl carbinol are not formed. Hydrogen sulfide is produced, and nitrates are reduced.

The organism can be cultivated on special media such as dextrose-lactose agar (Mallmann and Snyder, 1929), brilliant green agar (Mallmann, 1929), Endo's agar, cysteine gelatin (Hinshaw, 1941), and sodium tartrate and mucate media (Mallmann, 1931) which may be of value in the differentiation from other organisms. Bushnell and Porter (1945) tested several types of media for the cultivation and isolation of *S. pullorum*. They concluded that no single medium used proved satisfactory for isolation of *S. pullorum*. In the selection of the medium for the isolation of *S. pullorum* consideration must be given to the source of material to be examined. In isolating *S. pullorum* from the intestine desoxycholate citrate, bismuth sulfite, and S S agar were found to be the most satisfactory. Tetrathionate broth was recommended as an enrichment medium.

Reference to the literature (Hinshaw, 1941; Pacheco and Rodrigues, 1936; and Rettger and Plastridge, 1932) reveals that a disagreement con-

cerning the results of the biochemical behavior of S. *pullorum* is to be found; especially is this true among European and American workers. The former for the most part are inclined to regard S. *pullorum* and S. *gallinarum* as identical species. This is difficult to comprehend when certain areas in the United States are observed to be relatively free of fowl typhoid as based on field and laboratory observations, whereas pullorum disease is more prevalent. This statement is based on diagnoses obtained by the standard criteria employed in differentiating these two diseases.

While S. *pullorum* is generally accepted as being a stable distinct species, Plastridge and Rettger (1930, 1932), Mallmann (1932), and Van Roekel (1935) have observed that the organism may vary markedly in many of its characteristics (Fig. 8.2—5 to 8). The toxicogenic properties of S. *pullorum* were investigated by Hanks and Rettger (1932), who observed S. *pullorum* cells contained an extractable heat-resistant poison which is highly toxic for rabbits and is capable of killing guinea pigs and mice. Chicks revealed no noticeable symptoms of illness, regardless of the route by which the material was introduced. They concluded that pullorum disease appears to be a septicemia rather than a toxemia.

The antigenic composition of S. *pullorum*, according to the Kauffmann-White schema (Salmonella Subcommittee, 1934), consists only of the O-antigen. Its antigenic structure is similar to that of S. *gallinarum*. However, the O-antigen of S. *pullorum* and S. *gallinarum* has something in common with the somatic antigen of the other members in the Salmonella genus.

Edwards and Bruner (1946) in their study regarding the antigenic components of S. *pullorum* conclude the following: "The antigenic formula of S. *pullorum* is IX, $XII_1$, [$XII_2$], $XII_3$. In normal cultures the $XII_2$ factor is variable, and forms containing a large amount or a negligible amount of $XII_2$ can be isolated from the same strain. It is possible for cultures to become fairly well stabilized in either form, thus giving rise to the so-called "standard" strains and "variant" or X strains. The standard strains contain only a small amount of $XII_2$, but the X strains contain a large amount of the antigen."

## PATHOGENICITY

Pullorum infection is most prevalent among chickens. However, among the various poultry breeds, a difference in the susceptibility to S. *pullorum* may be apparent. The lighter breeds of chickens. especially the Leghorns, generally speaking, have revealed fewer reactors among tested flocks. Hutt and Scholes (1941) claim the Leghorns possess a greater genetic resistance to the disease. This view is in part also subscribed to by Roberts and Card (1935), although they state that strain differences within the various breeds
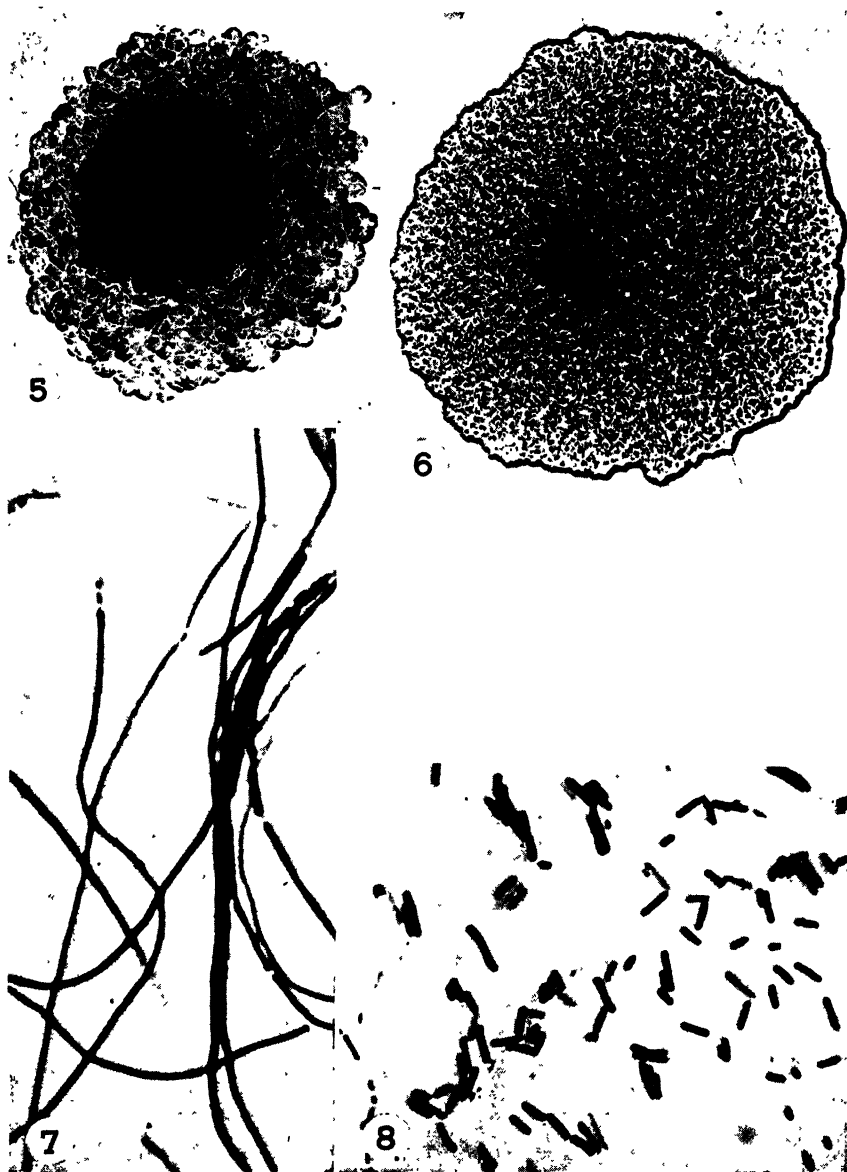
FIG. 8.2. 5—Colony exhibiting dentated edge, thin periphery which is markedly convoluted, striated, and tenacious. Central portion raised, dense, and faintly convoluted. Three days old. Liver infusion agar. ×10. 6—Colony exhibiting irregular outline, rough surface, opaqueness, and brittleness. Two days old. Meat extract agar. ×25. 7—Filamentous forms in smear prepared from the peripheral portion of the colony illustrated in 5. ×1,200. 8—Cells in a smear prepared from the central portion of the colony illustrated in 5. ×1,200.

must be considered. If Leghorns are to be regarded more resistant to pullorum disease on the basis of blood-testing results, then males likewise must be considered more resistant than females. Testing results for a period of years reveal a greater percentage of reactors among females than among males. This difference certainly cannot be attributed entirely to a hereditary trait existing in the male sex because the influence of environmental factors operative in a commercial plant should be recognized. From a series of investigations, Roberts and Card (1935) concluded that heredity is an important factor in resistance and susceptibility to infection with *S. pullorum*. Later Roberts, Severens, and Card (1939b) reported that resistance and susceptibility of the domestic fowl to pullorum disease are related to the number of lymphocytes. Resistant chicks revealed a higher lymphocytic count than did the susceptibles. Change in age of the chick also influenced the degree of resistance as well as the lymphocyte number. Scholes and Hutt (1942) claim that high body temperatures and resistance to *S. pullorum* are closely associated. Later Scholes (1942) concluded that resistance to *S. pullorum* more likely depends upon temperature differences than upon differences in the number of lymphocytes in the blood. While the observations reported by Hutt and Scholes (1941), Scholes (1942), Scholes and Hutt (1942), and Roberts *et al.* (1935, 1939b) command academic interest, from a practical viewpoint the combat against the disease through genetic selection of resistant stock does not appear as effective as the method of eliminating the disease carriers from the breeding flocks. This is substantiated by DeVolt, Quigley, and Byerly (1941) who state that the development of pullorum-resistant strains is not now a satisfactory substitute for control and eradication programs by the agglutination tests.

While the chicken appears to be the natural host of *S. pullorum,* other avian species also have exhibited some degree of susceptibility. Among the barnyard fowl, natural infection has been observed in turkeys (Barboni, 1937; Brunett, 1930; Dalling *et al.,* 1929; Hendrickson and Hilbert, 1930; Hewitt, 1928; and Hinshaw, 1937); ducks (Beaudette, 1938; Hinshaw and Hoffman, 1937; Lerche, 1929; and Miessner, 1931); and guinea fowl (Bunyea, 1939). Natural outbreaks among pheasants (Hendrickson and Hilbert, 1931; Miessner, 1931), quail (Emmel, 1936), sparrows (Dalling *et al.,* 1928; Reis and Nobrega, 1936), European bullfinch (*Pyrrhula europa*) (Hudson and Beaudette, 1929), and pigeons (Reis and Nobrega, 1936; van Heelsbergen, 1929) have also been reported. Canaries, goslings, turtle doves, gold finches, green finches, and bittern are reported vulnerable to infection (Villani, 1937). Edwards (1945) reported a loss of 50 birds among a flock of 75 canaries caused by a natural outbreak of pullorum disease. Thirteen birds were examined and *S. pullorum* was recovered from all of them.

Throughout the United States, in some sections more than in others,

pullorum disease has gained a considerable foothold in commercial turkey flocks (Hinshaw, 1937, 1939). The clinical and pathological aspects of the infection in the turkey are similar to those in the chicken. The natural incidence of pullorum infection among commercial turkey flocks is of great concern to those engaged in the control and eradication of pullorum disease among domestic fowl. A more detailed discussion of the disease among turkeys is presented in the section entitled Diseases of the Turkey.

Among other fowl which were found to be susceptible either through natural avenues or through artificial means, the disease produced clinical manifestations quite similar to those observed in the chicken and turkey (Dalling et al., 1928; Emmel, 1936; van Heelsbergen, 1929; Van Roekel et al., 1932). Pigeons apparently are quite resistant to the organism, whereas sparrows appear to be very susceptible. The fact that sparrows and pigeons may frequently inhabit poultry plants may offer an explanation why infection appears in previously nonreacting flocks.

Upland game birds such as pheasants and quail exhibit a degree of susceptibility to the extent that game breeders must exercise preventive measures against the disease.

Guinea fowl, ducks, and geese appear quite resistant to the organism, but their role in contracting and disseminating the infection should not be considered negligible in the combat against the disease.

Among the mammalian species the rabbit is highly susceptible, as observed by Olney (1928) and Doyle (1925). Guinea pigs, mice, and cats were slightly susceptible, while rats were quite resistant (Mulsow, 1919; Rettger et al., 1916). Benedict, McCoy, and Wisnicky (1941) recovered the organism from foxes and mink. Edwards and Bruner (1943) isolated a culture of porcine origin. Within recent years S. pullorum infection has been reported in man by Edwards and Bruner (1943), Felsenfeld and Young (1944), and Mitchell, Garlock, and Broh-Kahn (1946).

## MODES OF TRANSMISSION

The manner in which pullorum infection may be disseminated is of great importance from the standpoint of control, eradication, and prevention of the disease. The etiological agent may be spread through various channels (Fig. 8.3).

Rettger and Stoneburn (1909) isolated the organism from fresh and incubated eggs which were laid by hens whose progeny succumbed to the disease. Later investigations (Dearstyne, et al., 1929; Gage, et al., 1914; Jones, 1913a; Rettger and Stoneburn, 1911; Runnels and Van Roekel, 1927a, 1927b; Tittsler, et al., 1928) revealed that S. pullorum could be readily recovered from eggs. Runnells and Van Roekel (1927b) reported 33.7 per cent of the eggs laid by reacting hens to be infective. Jones (1913a) and Mathews (1927) reported outbreaks in adult fowls due to S. pullorum as the

result of feeding incubated eggs. Van Heelsbergen (1929) emphasized that an important channel of pullorum disease dissemination is through so-called "egg-picking." Rettger (1916) stated that eggs harboring large numbers of the organism produce abnormal conditions when fed to young chicks, adult fowls, young rabbits, guinea pigs, and kittens. Olney (1928) encountered a severe outbreak of the disease among adult rabbits as a result of feeding
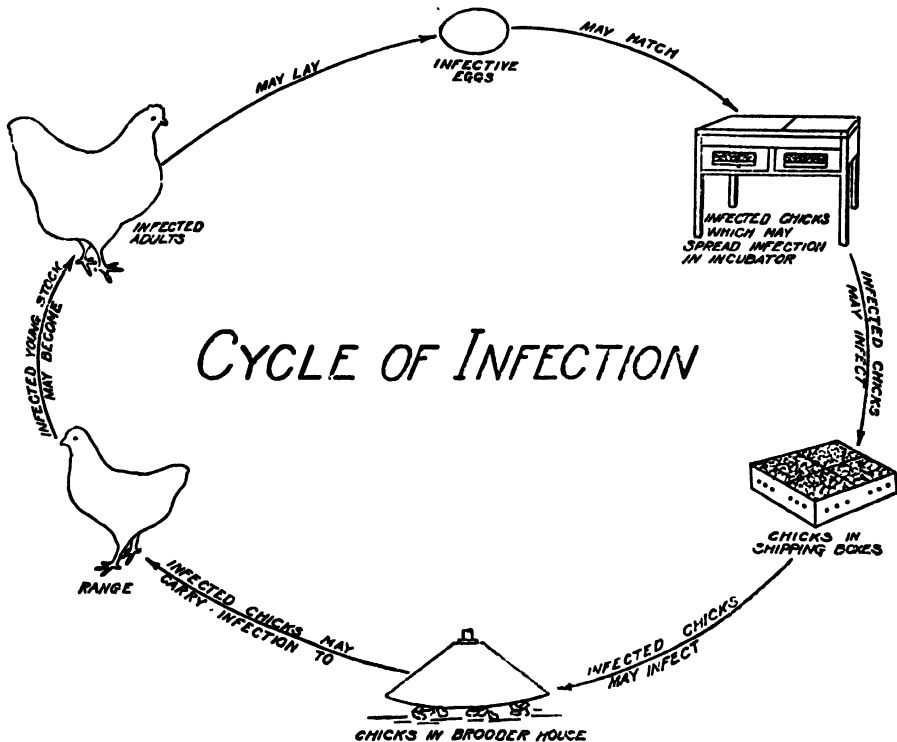


FIG. 8.3. Pullorum infection in a flock may follow this cycle.

incubated, infertile eggs. Van Roekel, Bullis, Flint, and Clarke (1932) reported that fresh eggs laid by reacting hens may produce pullorum disease when fed to noninfected hens and pullets, and it appeared that younger birds may contract the disease more readily through eating infective eggs than do older birds. They concluded that the habit of "egg-eating" or "egg-picking" in an infected flock should be regarded as a hazard to an eradication program for such a flock.

Flock conditions may increase the spread of the disease by means of the egg. Inadequate nesting facilities, which may cause birds to crowd in the nest or lay their eggs on the dropping boards and floor, result in an increase in the number of broken eggs. Pullets reaching sexual maturity may frequently lay soft-shelled and hard-shelled eggs on the floor and dropping

boards. The production of thin-shelled eggs may follow outbreaks of certain diseases. All these factors are of great significance in the spread of pullorum infection in an adult flock.

The excreta of infected birds must be considered a means by which infection may spread to noninfected birds in a flock and also from one farm to another. Kerr (1930) and Van Roekel, Bullis, Flint, and Clarke (1935) have reported the recovery of the organism from the feces of hens. Van Roekel, *et al.*, (1932, 1935) observed that fecal transmission of the disease among semimature and adult stock apparently occurs very infrequently. Transmission was observed only after force-feeding repeated doses of feces from infected hens. The dissemination of the disease in an adult flock by means of feces or litter contaminated with *S. pullorum* may be influenced by the numbers of organisms eliminated and the persistency of such elimination together with suitable environmental conditions. Such factors constitute a major problem in the eradication of the disease in a short interval retesting or multiple testing program of an infected flock.

Furthermore, cannibalism in an infected flock may further the spread of the disease. The abdominal viscera of infected birds in many instances are heavily contaminated with *S. pullorum,* and when such birds are eviscerated through cannibalistic habits in a flock, they may serve as a source of infection for other birds in the flock.

The important discovery of the organism in the egg established one phase in the cycle of infection. The most frequent spread of the disease occurs from the breeding female to its progeny by way of the egg. At the present time, this is the most common mode of transmission and will continue to be the case if infected birds are tolerated in a breeding flock. It is recognized that the greater the number of infected birds in a breeding flock, the greater will be the number of infected chicks. It has also been observed that one or two infected breeding birds may be responsible for serious infection in the progeny.

Transmission of the disease in incubators through chick excretions, egg shells, and chick down has been recognized as a very serious problem in the control of the disease for many years. Hinshaw, Upp, and Moore (1926) definitely revealed that artificially contaminated chick down could disseminate the disease in a forced-air-draft-type incubator. It was emphasized that in commercial hatching, precautions should be exercised against the spread of the infection through the incubator. Bunyea and Hall (1929) pointed out that pulmonary and cardiac lesions appear to represent a form of the disease acquired by inhalation and that gross intestinal lesions, such as thickening and necrosis of the large intestine, are indicative of infection by ingestion. The significance of incubator transmission of the disease will be discussed later in this chapter.

From the time the chick is removed from the incubator to the time it is placed in a brooder, it may contract the infection through being handled or through contact with infected chicks in the same chick box. Droppings of infected chicks serve as the source of the infection. Chick boxes and any other equipment of a like nature should not be used a second time unless properly cleaned and disinfected. Chick handlers, and especially chick sexers, should give consideration toward reducing the spread of the infection when chicks from different flocks are handled.

The infection, as a rule, is spread among chicks in cohabitation during the brooding stage, especially during the first few days of age. According to Weldin and Weaver (1930), the chief source of the organism at this stage is the droppings. Mallmann (1929), through the use of a brilliant green enrichment medium, was able to isolate the organism from the feces of infected chicks. He observed that when S. *pullorum* was in the organs of the chicks, it was nearly always found in the intestinal contents. Litter, feed, and water soiled with infective droppings aid in the rapid spread of the infection in a chick flock. Gwatkin and Mitchell (1944) found that pullorum disease could be produced in chicks which had access to feed contaminated by infected flies and to the flies themselves, some of which were probably eaten by the chicks. S. *pullorum* was recovered from the feet and wings of flies at least six hours after exposure. The gastro-intestinal tract of the flies was found to harbor the infection for at least 5 days.

The mode of spread among young chicks is often facilitated through environmental factors such as extremes in temperature, insanitary conditions, lack of or inadequate feed, and other diseases appearing concurrently. It is frequently observed that chicks which originate from a hatchery not recognized as free from pullorum infection and which are subjected to unfavorable conditions in transit will manifest greater evidence of the disease than will chicks from the same source which are subjected to more favorable conditions. Proper brooder management plays a very important role in keeping the spread of infection down to a minimum.

Pullorum infection is likely to occur regardless of the portal of entry. Van Roekel, Bullis, Flint, and Clarke (1932) report that pullorum disease can be reproduced in chickens by dropping suspensions of the organism on the conjunctiva, into an incision in the skin of the plantar surface of the foot, into the cloaca, and by oral administration. However, the oral route did not yield to the establishment of infection as readily as others that were investigated. The presence of agglutinins was detected 6 days after exposure in the case of cloacal inoculation, 7 days with the ocular route, 10 days each for the oral route and skin incision instillation. In some cases, agglutinins were produced but later disappeared from the blood stream. Likewise, this might occur in a naturally infected flock. Some investigators (Bunyea and Hall,

1929; Doyle and Mathews, 1928; Hinshaw, Upp, and Moore, 1926) reported
that infection may result from the entrance of the organism into the respira-
tory and alimentary tracts. Apparently the portal of entry for post-hatching
infection is more often the digestive tract than the respiratory tract.

Hence, it appears that *S. pullorum* may be eliminated and excreted from
the infected host in various ways and in turn may enter the body through
various avenues when suitable environment exists. This is significant from
the standpoint of control, eradication, and prevention of the disease.

### SYMPTOMS AND LESIONS

*Adult fowl.* Pullorum disease in a maturing or an adult flock does not
manifest the characteristics of an acute infection as a rule. The spread of
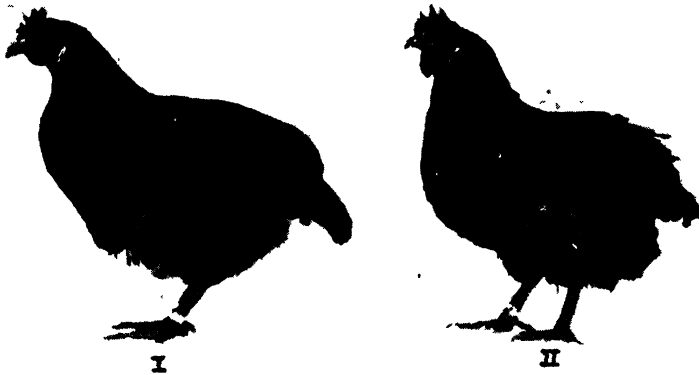


FIG. 8.4. Bird No. I—noninfected hen. Bird No. II—infected hen.

infection within a flock may occur continuously, but the flock owner may not
be aware of it. In contracting the infection, the bird may exhibit little or
no symptomatology. Infected individuals cannot, as a rule, be detected by
their physical appearance (Fig. 8.4). Experimentally infected birds have
exhibited limited and transient clinical manifestations. A general depres-
sion and listlessness, accompanied by a partial or complete inappetence, may
be the first symptoms following the infection. A paleness of the comb and
visible mucous membranes may be observed. Diarrhea may be noted. A
febrile reaction accompanied by increased thirst has been observed. Occa-
sionally adults may succumb to artificial infection depending upon the
dosage and virulency of the organism.

Natural epornitics in a flock have been observed. Jones (1913a) reported
a natural outbreak among adult fowl which was attributed to the feeding of
eggs discarded from an incubator. Evidence of disease was observed 16 days
after the feeding of the infective eggs, and during a period of six weeks a loss

of 50 birds among a flock of 700 hens was sustained. The symptoms noted were paleness of the comb and mucous membranes; scaly, shrunken, and grayish appearance of the comb; listlessness; progressive depression; droopy wings, retraction of head and neck; inappetence; and usually diarrhea. The duration of the incubation period was from 16 days to three weeks. The course of the disease sometimes terminated fatally in 24 hours, usually continued 4 or 5 days, and occasionally even longer. A definite leukocytosis was observed.

Plastridge and Rettger (1930) have observed among adult flocks acute outbreaks of the disease caused by a highly pleomorphic type of *Salmonella pullorum*.

Among birds dying from acute infection, Jones (1913a) observed marked emaciation; enlarged and distorted heart due to grayish-white nodules; enlarged, yellowish-green and granular liver coated with fibrinous exudate; friable spleen of normal size with focal necrosis; minute necrotic foci of the pancreas; enlarged kidneys with parenchymatous degeneration; injection of mesenteric vessels and a fibrinous exudate coating the abdominal viscera.

The lesions found in the more common chronic carrier are the misshapen, discolored, cystic ova (Fig. 8.6), and frequently an acute or chronic pericarditis. The diseased ova usually contain oily and cheesy materials enclosed in a thickened capsule. The organism can be readily isolated from the ovarian cysts. These cysts may be closely attached to the ovary, but frequently they are pedunculated and may become detached from the ovarian mass. In such cases, they become embedded in the adipose tissue of the abdominal cavity. In one case, a pedunculated ovum was recovered as a foreign body from a normal-appearing egg (Fig. 8.5). Ovarian and oviduct dysfunction may lead to abdominal ovulation or oviduct impaction, which in turn may bring about extensive peritonitis and adhesions of the abdominal viscera (Fig. 8.7). Advanced lesions of this type seldom, if ever, fail to yield *S. pullorum* on culture.

Lesions less extensive in nature may involve the heart. Quite frequently pericarditis is observed both among females and males. The changes that have occurred in the pericardium, epicardium, and pericardial fluid appear to be dependent on the age of the disease process. In some cases, the pericardium exhibits only a slight translucency, and the pericardial fluid may be increased and possess a turbidity. In the more progressive stages, the pericardial sac is thickened and opaque, and the pericardial fluid is greatly increased in amount, containing considerable exudative material. This may be followed by permanent thickening of the pericardium and epicardium and partial obliteration of the pericardial cavity by adhesions (Fig. 8.8). The organism can usually be recovered from such a process.

In the male the infection is frequently found in the reproductive organs.

Edwards and Hull (1929) reported the localization of the organism in the testicle and vas deferens. A thickening of the tunica albuginea and complete obliteration of the seminiferous tubules were observed. The testis revealed multiple small abscesses and areas of round cell infiltration. There was no evidence of spermatogenesis. The lumen of the vas deferens was enlarged



Fɪɢ. 8.5. Pedunculated ovum obtained from an infertile egg containing an apparently normal yolk. *S. pullorum* was isolated from its contents. ×2.

and filled with a dense structureless homogeneous exudate. Other cases of infection in the male reproductive system have been reported (Fig. 8.9). Pericarditis is frequently observed among infected males, and less frequently small infective cysts containing amber-colored, cheesy material may be found embedded in the abdominal fat or attached to the gizzard or intestines.

Bushnell, Hinshaw, and Payne (1926) reported the isolation of the organism from abscesses on the skin and the legs. Subcutaneous abscesses over the sternal region and cystic enlarged thyroids have yielded the organism. Gwatkin and Glover (1930) isolated the organism from the nasal passages of two among sixty-one adult birds examined. Beach and Freeborn (1927) reported the isolation of *S. pullorum* from the middle ear with no
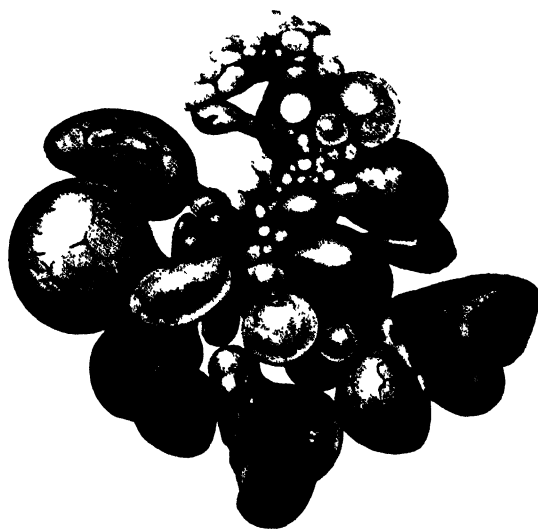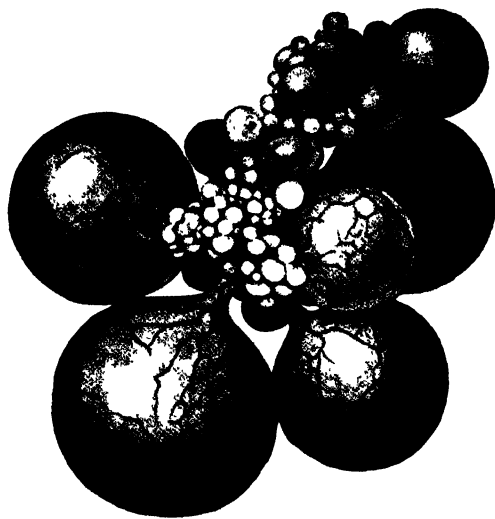
FIG. 8.6. Above—normal ovary. Below—infected ovary (*S. pullorum*). (Storrs Agr. Exper. Sta.)

evidence of infection in any other part of the body. Gwatkin (1946) recovered the organism from the thymus glands.

*Chick.* Manifestations of pullorum disease were first recognized among young chicks, and the malady may be considered as principally a chick disease. The symptoms exhibited by an infected brood of chicks are not



FIG. 8.7. Impacted oviduct removed from an infected hen. Parts I and II are the funnel and albumen secreting portions, respectively. Part III is the shell gland portion. Adhesions of the serosa.

specific for pullorum disease, although in many cases a tentative diagnosis based on clinical evidence has been substantiated by the isolation of the etiological agent.

In a typical outbreak of infection among chicks, the following symptoms may be observed: The onset of the disease varies with the degree, virulence,

and source of the infection, and with the management given the chicks. If chicks are hatched from infective eggs, morbid and dead chicks may be observed in the incubator or within a short time after hatching. The chicks manifest a somnolence, weakness, and loss of appetite, and death may follow suddenly. In some instances, evidence of the disease is not observed until several days (5 to 10) after hatching. The disease gains momentum during the following 7 or 10 days. The peak of the infection usually occurs during



FIG. 8.8. Left—normal heart. Right—infected heart exhibiting pericarditis and epicarditis.

the second or third week of chickhood. In such instances, the chicks exhibit a lassitude, an inclination to huddle together under the hover, loss of appetite, an accumulation of urinary and alimentary excretions in and around the vent, drooping of the wings, somnolence, and a distorted body appearance (Fig. 8.10). Frequently one may detect a shrill cry from a chick when voiding excreta. This is particularly true among those chicks that have an accumulation of whitish, chalk-like excreta, stained greenish-brown, in and around the vent.

Jungherr (1935) reports that affected chicks manifest a febrile reaction as indicated by the increased temperature of the legs. He also mentions that affected chicks may appear as having been "dipped in water" which he explained as probably brought on by a water-logged condition of the body

muscles which permits the excess fluid to ooze through the skin. Hutyra, Marek, and Manninger (1938) claim that a febrile condition is responsible for the increased renal activity and elimination of the whitish material adhering to the vent and adjacent parts.

Labored breathing may be observed even to the extent that chicks may be gasping for breath. This should not be confused with what is commonly designated "brooder pneumonia" or mold infection, and neither should it be mistaken for infectious bronchitis, Newcastle disease, or some other respiratory disturbance. The mortality may vary from no losses to approximately 100 per cent in serious outbreaks. The morbidity and mortality rates are dependent upon many factors. The greatest losses occur usually during the second week after hatching with a rapid decline during the third and fourth weeks. Survivors may be greatly retarded in their growth and appear as underdeveloped and poorly feathered birds (Fig. 8.11). However, some survivors may not reveal any great setback in growth, but grow to maturity even though har-



FIG. 8.9. Testicles removed from a reacting adult male. Testis I—atrophic and very firm; *S. pullorum* isolated. Testis II—normal in size and texture; *S. pullorum* not isolated.

boring the infection. Chick flocks which have passed through a serious outbreak usually reveal a high percentage of carriers at maturity.

Chicks hatched from an infected flock and raised on the same premises will usually reveal less mortality from the disease than chicks from the same flock shipped away.

In chicks that die suddenly in the early stages of brooding, the lesions are limited. The liver is enlarged and congested, and the normal yellow color may be streaked with hemorrhages. In the septicemic form, an active hyperemia may be found in other organs. The yolk sac and its contents

reveal slight or no alteration. In the more protracted cases, an interference with yolk absorption may occur, and the yolk sac contents may be yellowish in color and of creamy and cheesy consistency. Necrotic foci or nodules may be present in the cardiac muscle (Fig. 8.12), liver, lungs (Fig. 8.13), ceca, large intestine, and the muscle of the gizzard. Pericarditis may be observed
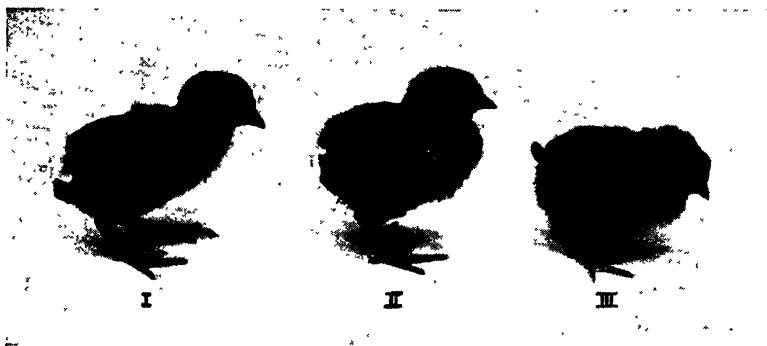


Fig. 8.10. Nine-day-old, naturally infected pullorum disease chicks. *S. pullorum* was isolated from chicks II and III. Chick III died two days after it was photographed.
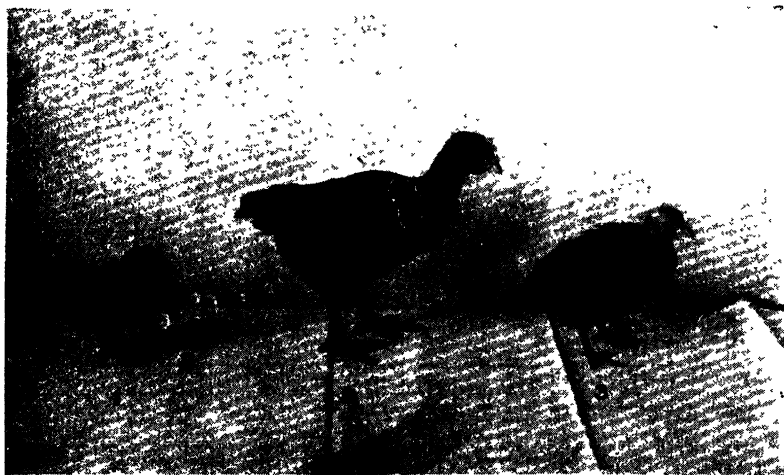


Fig. 8.11. Six-weeks-old chicks exposed to pullorum infection when 72 hours old. Weights: No. 1, 115 gm.; No. 2, 488 gm.; No. 3, 193 gm.

in certain instances. The liver may reveal punctiform hemorrhages and focal necrosis. The spleen may be enlarged (Fig. 8.14) and the kidneys congested or anemic with ureters prominently distended with urates. The ceca may contain a cheesy core sometimes tinted with blood which should not be confused with a somewhat similar lesion encountered in coccidiosis. The wall of the large intestine may be definitely thickened. Frequently peri-

tonitis is manifested. Doyle and Mathews (1928) report that the liver is the most constant seat of gross lesions and followed in order by the lungs, heart, gizzard, and ceca. Among chicks only a few days old, the lung lesions may consist only of a hemorrhagic pneumonia, whereas in older chicks small yellowish-gray nodules and areas of gray hepatization may be found. The nodules in the myocardium may attain a size causing a marked distortion in the shape of the heart.

The histopathologic findings in young chicks affected with pullorum disease present features not markedly different from those in other infectious diseases. Doyle and Mathews (1928) state that in young chicks the livers show hyperemia, hemorrhages, focal degeneration, and necrosis (Fig. 8.15).



FIG. 8.12. A—pullorum diseased heart exhibiting nodular abscesses in the myocardium (16-day-old chick). B—normal heart (17-day-old chick).

They claim that the accumulation of endothelial leukocytes which replace the degenerated or necrotic liver cells is a characteristic cell reaction of the liver to *S. pullorum* infection. The histopathologic pulmonary lesions may consist of diffuse, acute congestion and hemorrhage in the early stages. Later, well-defined focal lesions appear which consist chiefly of a mononuclear infiltrating type of cell, serofibrinous exudate, and cellular debris. The



FIG. 8.13. No. 1 from left—normal lung (17-day-old chick). Nos. 2, 3, and 4—pullorum infected lungs exhibiting pneumonia and multiple abscesses (16-day-old chick).

larger lesions may involve several lobules, bronchioles, and bronchi terminating in necrosis.

The nodules in the myocardium and in the muscle of the gizzard represent largely an infiltration with mononuclear cells and degenerative changes of the muscle fibers.

The pulmonary and cardiac lesions may be considered of considerable diagnostic importance. However, for an accurate diagnosis of diseased speci-

mens, the presence or absence of these findings should be confirmed by bacteriological examination.

## DIAGNOSIS

The identification of pullorum disease in maturing and adult stock should be made on the basis of serological findings and not on clinical and post-mortem observations. Bacteriological examination should be made in acute cases of the disease and in certain instances when the serological findings may appear questionable. Other infectious diseases such as fowl typhoid, fowl cholera, paratyphoid infection, and "unknown disease" may at times be difficult to differentiate from pullorum infection without recourse to a bacteriological examination. Disturbances of the female reproductive system are common in chickens, but pullorum disease can be incriminated for only a portion of them



FIG. 8.14. A—normal spleen (17-day-old chick). B—pullorum infected spleen exhibiting marked enlargement (16-day-old chick).

(Fig. 8.16). Serological findings should not be considered final for all suspicious cases of pullorum disease. Fowl typhoid infected birds and occasionally those harboring paratyphoid organisms will give a positive test when their sera are tested with pullorum antigen. Furthermore, it has
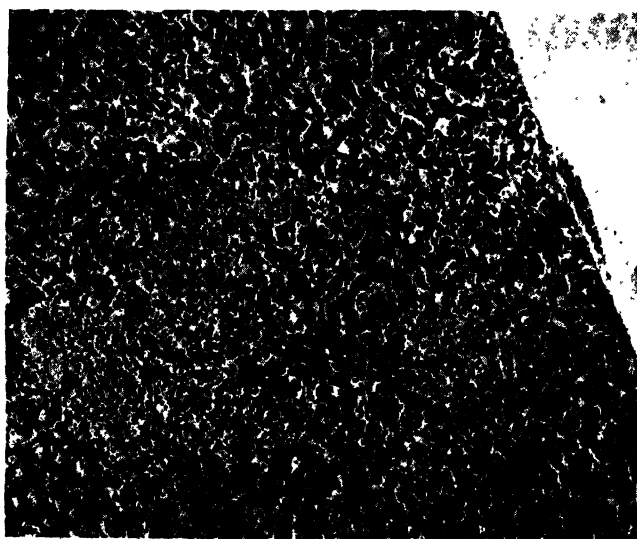


FIG. 8.15. Liver revealing focal degeneration and necrosis. ×100.

been observed in routine pullorum testing that other bacterial organisms may cause birds to produce sera that will give nonspecific reactions with pullorum antigen (Garrard, Burton, and Carpenter, 1947; Gwatkin, 1946).

Bacteriological examination should be supplemented in such instances for an accurate diagnosis.

The diagnosis of the disease in young chicks should be based on bacteriological examination even though well-pronounced lesions which may be considered quite characteristic of the infection are observed. Such a diagnostic procedure should definitely determine the presence or absence of fowl typhoid, paratyphoid, staphylococcosis, coccidiosis, infectious bronchitis, aspergillosis, and noninfectious diseases.

The Standard Methods of Diagnosis of Pullorum Disease in Barnyard Fowl (Anon., 1933) which were adopted by the Conference of Official State and Federal Research Workers in Animal Diseases of America in 1931 and by the United States Livestock Sanitary Association in 1932, state that "the only criterion of infection with *Salmonella pullorum* shall be the isolation of this organism from the blood and body tissues of suspected chicks and its complete identification." Ordinary standard beef infusion agar is recommended for the isolation of the organism, and in case chicks have been in transit for some time leading to partial decomposition of the specimens, brilliant green liver infusion medium as described by Mallmann (1929) is recommended.



FIG. 8.16. Cystic ovary infected with *Escherichia coli*.

Cultures should be incubated from 24 to 48 hours at 37° C., and characteristic colonies should be identified by Gram-stained slide mounts and by their fermentative reaction in glucose, lactose, saccharose, and dulcitol. The antigenic specificity should be tested against known positive and negative pullorum sera. A Gram-negative medium rod, producing acid and gas, or acid in only glucose, and agglutinating with positive serum is to be regarded as a typical criterion for *Salmonella pullorum*. The antigenic specificity may be tested with the rapid serum method as recommended by Stafseth and Corbutt (1940) which may expedite the completion of the examination and the reporting of the diagnosis to the consignee. The time usually required to obtain a diagnosis is approximately 48 to 96 hours after the consignment arrives at the laboratory. Reliable and expeditious diagnostic service is very essential because it may avoid serious losses and spread of the disease.

### THERAPEUTICS

Until recently efforts to reduce losses in outbreaks of pullorum disease through medicinal treatment have met with little or no success. Drugs and chemicals including chinosol, metaphen, sulfuric acid, hydrochloric acid, mercuric chloride, resorcin, potassium permanganate, sulfocarbolates, and hypochlorite solutions have been reported to have no beneficial influence when taken into the alimentary tract (Beach and Freeborn, 1927). The hypochlorite solutions were found to be of some value as a drinking water disinfectant. However, from a practical standpoint, considering the various other channels through which the organism may spread in a flock, it appears doubtful whether the expenditure for a drinking water disinfectant would be justified. Recent introduction of sulfonamides in the control of the infection has yielded promising results (Severens, Roberts, and Card, 1945; Bottorff and Kiser, 1946; Gwatkin, 1946). However, further investigational work seems apparent to ascertain their practical value in the control of the disease in acute natural outbreaks. The use of sanitary drinking fountains and feed hoppers, the frequent removal of contaminated litter, the maintenance of proper and uniform brooding temperatures, the avoidance of overcrowding, and the prompt removal of visibly sick and dead chicks are management measures which will reduce the spread of the infection and minimize losses.

Moore, Mallmann, and Arnold (1934) have shown that increasing the brooder temperature from 5 to 10 degrees above that recommended for normal brooding operations will reduce the mortality. Bushnell, Hinshaw, and Payne (1926) concluded that the feeding of sour milk is of little value in the control of pullorum infection, except that it may increase the vigor of the chicks due to its food value. Roberts, Severens, and Card (1939a) reported that diet exerted an influence on the morbidity and mortality due to S. pullorum. A high mortality resulted from the feeding of laying mash, but when chick mash was substituted the high mortality disappeared.

Biotherapy (vaccines, bacterins, and serums) (van Heelsbergen, 1929) has been attempted but has not given satisfactory results. According to Mallmann (1931a), phage therapy also has proved to be ineffective in the control of the disease.

Incubator sanitation and disinfection are very essential in combating pullorum disease. At the start of hatching operations and repeated between hatches for the duration of the hatching season, incubators should be thoroughly cleaned and disinfected. Hatcherymen frequently overlook the fact that thorough cleaning should always precede disinfection. Liquid disinfectants and fumigants have their effectiveness greatly reduced if used in incubators containing chick down, egg shells, excreta, and other debris.

Formaldehyde, an extensively used fumigant, has been found very effec-

tive in incubator disinfection. Investigations (Bushnell, Payne, and Coon, 1929; Bushnell and Payne, 1931; Graham, 1941; Gwatkin, 1927; Insko, Steele, and Hinton, 1941) have revealed that definite procedures must be followed to obtain the optimum results. It is generally recommended that 35 cc. of formalin be added to 17.5 grams of potassium permanganate to fumigate 100 cubic feet of incubator space with wet-bulb and dry-bulb temperature readings of 86–90° F. and 100° F., respectively. A more recent report (Burton, 1946) emphasizes that at least 150 cubic centimeters of formalin and 100 grams of potassium permanganate be used to fumigate 100 cubic feet of inside incubator space. After 20 minutes exposure to the gas, complete destruction of S. *pullorum* was observed. It is stressed that factors such as air leakage, improper humidity and temperature, circulation of gas within the incubator, and duration time of fumigation all play a very important role in effective fumigation of the incubator. Considerable leakage of gas was found in some commercial machines. Earlier Graham (1941) reported that maximum results of fumigation (using 35 cc. of formalin and 17.5 grams of potassium permanganate for each 100 cubic feet) may be expected only if incubators are relatively clean and if the doors of forced-draft incubators are kept closed for a minimum of 3 hours following the release of the formaldehyde. The gas may then be liberated from the machine either by opening the doors for a few minutes or by neutralizing with strong ammonia water. The ammonia water may be sprinkled on the walls of the inside chamber. In forced-draft incubators, the diffusion of gases is very rapid and effective; however, first the proper temperature and humidity should be established. Insko, Steele, and Hinton (1941) advise raising the wet-bulb reading to 92–94° F. at the time of fumigation and maintaining the dry-bulb thermometer at normal operating temperature. Fumigation at high concentrations during the first 3 days of incubation is advised against, because the embryos at this period can tolerate less formaldehyde than at a later age. Losses of practical significance were not observed until four times normal concentration (35 cc. formalin per 100 cubic feet of space) of the fumigant was used.

When eggs are hatched in separate hatching compartments, they should be fumigated on the eighteenth to twentieth day of incubation. Fumigation may be repeated at short intervals during the hatching process but should not be delayed until the chicks have dried. The relative humidity should be maintained at a high level which will aid the chick in withstanding the gas. The incubator should remain closed for 8 to 10 minutes. Immediately after each fumigation all chicks, whether wet or dry, should be removed from the incubator and kept in a comfortable environment. Ammonium hydroxide may be used to advantage in neutralizing the formaldehyde and in facilitating the handling of the chicks.

In addition to the formalin and potassium permanganate method, the use of cheesecloth saturated with formalin has been recommended. This latter method may be more economical in the use of formalin, but it requires a longer fumigation period. Commercial fumigants (Graham, 1941) are also available on the market, but the public should refrain from using such preparations until they are endorsed by qualified authorities.

Effective incubator fumigation by the formaldehyde method cannot be expected unless proper conditions prevail, and even then certain limitations exist as in the case of fumigating chicks in the process of hatching. Complete destruction of S. pullorum does not occur, but it is possible to reduce the chances of infection to some degree within the incubator. Incubator fumigation should be regarded as only one step or means that may be employed to advantage in a program designed for the control, eradication, and prevention of the disease.

### CONTROL AND ERADICATION

Rettger, Kirkpatrick, and Jones (1914) first reported that they had definitely established the complete cycle of infection and that chicks which were infected with the organism when small may develop into permanent carriers and be a constant source of danger to young and old stock. They found that the carrier condition might be established in approximately 25 per cent of an infected flock. To successfully combat the disease, these investigators stated that the infection cycle should be broken. Attempts were made to detect the carriers in flocks by the recognition of diseased ovaries in birds that were slaughtered for meat. This method was found inadequate and impractical in eliminating all infected birds from the flock.

A later method, the bacteriological examination of fresh eggs from an infected flock, was also found impractical, unreliable, and costly in the eradication of the disease.

Jones (1913b) reported the use of the macroscopic tube agglutination test as a means of detecting carriers of the infection and recommended the lower serum dilutions (1:50, 1:100, and 1:200) for routine testing (Fig. 8.17). The test was considered to be of great value in detecting infected fowl, but its practicability was dependent upon the value of the breeding flock.

The important contribution by Jones must be regarded as the foundation work for the diagnostic procedures which are in use at the present. Gage, Paige, and Hyland (1914), and Rettger, Kirkpatrick, and Jones (1915) substantiated the results of Jones and later employed the test in statewide campaigns with the objective of detecting flocks free from the disease as well as eliminating the disease by the detection and removal of the reactors.

In 1914 Rettger et al. (1915) tested 107 flocks representing 14,617 birds

which revealed 9.85 per cent reactors. Among 786 males tested only 2.9 per cent reacted. As to range of infection in terms of percentage, the tested flocks were classified as follows: 0 per cent, 28; 1–10 per cent, 34; 11–20 per cent, 18; 21–30 per cent, 10; 31–40 per cent, 10; 41–50 per cent, 4; 50+ per cent, 3. The White Leghorn flocks revealed a lower positive flock incidence than did the heavier breeds (57.1 per cent White Leghorns, 84.6 per cent Barred Rocks, and 88.4 per cent Rhode Island Reds).

The results obtained from the practical application of the macroscopic tube agglutination method revealed that a single test is not sufficient, as a rule, for the complete elimination of infected birds from a flock. However, improvement in hatchability and livability was observed among eggs and chicks, respectively, produced by tested flocks. It was recommended that only nonreacting birds be used for breeding purposes.

The initial testing of flocks in Massachusetts revealed results comparable to those reported in Connecticut. It is of interest to note that pullorum disease-free flocks did exist, while other flocks might reveal a large percentage of reactors.

As the testing programs expanded and additional



FIG. 8.17. Macroscopic tube agglutination test. Left—negative reaction. Right—positive reaction.

states adopted the routine testing of flocks, the test itself was gradually modified and improved as to its technique. Various investigators, organizations, and federal and state agencies have contributed to the serological diagnostic methods employed at the present time. The National Poultry Improvement Plan (Anon., 1941) endorses three methods [(1) The Standard Tube Agglutination Test; (2) The Stained-Antigen, Rapid Whole-Blood Test; and (3) The Rapid Serum Test] for official testing. The choice of method is determined by many factors and objectives peculiar to a state or a region. In some states the macroscopic tube agglutination method is considered the only official method for testing of flocks. In other states the rapid serum method

and the tube test are regarded as official. In many states all three methods may be employed, but the whole-blood test is the one most generally used.

The techniques and procedures for the three official methods are described in detail by the Bureau of Animal Industry, United States Department of Agriculture, Washington, D. C. (Anon., 1941). A brief review of each method is as follows:

### THE STANDARD TUBE AGGLUTINATION TEST

The blood samples shall be collected by a properly qualified and authorized person. Suitable blood tubes, shipping containers, and bleeding and



FIG. 8.18. Collecting blood samples for laboratory test.

leg-banding equipment should be furnished by the diagnostic laboratory or the authorized agency in charge of the testing program (Fig. 8.18). Blood tubes should be thoroughly cleaned and heated in a hot-air sterilizing oven. Cork stoppers should be boiled or washed and dried in a hot-air drying oven. Shipping containers for the blood samples should be constructed to permit washing and disinfection.

All birds tested are to be officially leg banded. The blood tube is identified with the leg-band number which is inscribed on the etched portion of the tube or on a gummed label. A small amount of blood (½ to 2 cc.) is collected from the median vein of the wing (Vena cutanea ulnaris) by

incising the latter with a sharp-pointed lancet or knife (Figs. 8.19 and 8.20).
The tube is laid on its side permitting the blood to clot in a long slant. After
the blood has coagulated, the samples are packed in containers designed for
shipment by mail, express service, or special messenger to the laboratory. In
extremes of temperature, precautions should be taken against freezing or
overheating because the blood samples should arrive at the laboratory in a
fresh state and unhemolyzed condition for a satisfactory test. All hemolyzed
or spoiled samples should be rejected. The diagnostic laboratory should be
equipped with proper and adequate refrigeration facilities where blood
samples should be retained until the sera have been tested and results of the



FIG. 8.19. Incising the median vein of the wing (Vena cutanea ulnaris).

tests are known. Occasionally, a retest on the same serum may be necessary
to determine the pullorum status of a bird.

The antigen for the tube test should be prepared from representative
strains of S. *pullorum* which are known to contain the different antigenic
components normally found in S. *pullorum* (Edwards and Bruner, 1946).
Furthermore, the strains should possess high agglutinability with positive
serum but should not agglutinate with negative or nonspecific sera. Stock
cultures of the antigen strains should be grown and maintained on nutrient
agar medium composed of dry granular agar (Difco) 2.0 per cent, Bacto
peptone (Difco) 1.0 per cent, beef extract (Difco) 0.4 per cent, and water.
The final hydrogen-ion concentration should range from 7.0 to 7.2. The

cultures should be transferred not more than once a month. Seed cultures should be taken from the stock strains rather than from rapid serial transfers in order to avoid contaminants or possible variation in the characteristics of the organism. Large test tubes, Kolle flasks, or Blake bottles containing nutrient agar medium may be used for producing the antigen. After 48 to 72 hours' incubation, the growth is washed off with sufficient phenolized (0.5 per cent) saline (0.85 per cent) solution to produce a very concentrated suspension. This suspension is filtered through sterile absorbent cotton or



FIG. 8.20. Collecting the blood into a numbered etched tube.

glass wool into sterile glass-stoppered bottles. The washings for each of the three strains are combined in equal volume-density, and the stock antigen is stored at 8–10° C.

For routine testing, a dilute antigen is prepared from the stock antigen by diluting the latter with physiological saline solution containing 0.25 to 0.3 per cent phenol. The turbidity of the antigen corresponds to 0.75–1.00 on the McFarland nephelometer scale, and the hydrogen-ion concentration is adjusted to pH 8.2–8.5 by the addition of dilute sodium hydroxide. The dilute antigen is prepared each day in order to reduce dissolution and plasmolysis to a minimum at the specified hydrogen-ion concentration.

The amount of diluted antigen employed in individual tests may vary from 1 to 2 cc.; however, the amounts should be constant and placed in clean, clear test tubes. Commercial devices are recommended for this phase of the work. The sera are added to the test tubes containing the antigen with a

serological pipette or a serum-delivery device which is accurately calibrated to deliver definite amounts. The antigen-serum mixture should be mixed by agitation and incubated for 24 hours at 37° C. and 24 hours at room temperature, depending upon the serum-antigen dilution. A 24-hour incubation period at 37° C. is sufficient and reliable when 1:25 dilution is employed. In the case of the 1:50 dilution, an additional 24 hours' incubation at room temperature appears necessary.

The results of the tests are interpreted as follows: *Negative test* represents a test in which the fluid remains uniformly turbid. *Positive test* represents a test in which the antigen reveals a distinct clumping, and clumps of cells have settled to the base of the tube with the supernatant fluid being clear. Gradation of clumping or agglutination may occur between negative and complete positive tests. These may be designated as slightly and strongly suspicious.

All suspicious and positive reacting tests should be reported to the agency responsible for the disposition of infected birds. Also, all broken, missing, and spoiled samples should be reported. In case the past status of the flock has been free of infection and only a few reactors are detected, the serological diagnosis should be confirmed by bacteriological examination of the reactors. Such a procedure will avoid a false diagnosis of fowl typhoid or paratyphoid infections. If only suspicious reactions are observed in a flock, then the strongest reacting birds should be submitted to the laboratory for retesting and a careful bacteriological examination. In routine testing, flocks should not be condemned as infected on the basis of doubtful or atypical reactions because such reactions may be due to causes aside from *S. pullorum*. If no conclusive evidence of pullorum infection can be found, the flock should be regarded as negative. This statement is based on observations made in routine testing in the New England States during the past decade (Van Roekel and Bullis, 1937). The lowering or removing of the official pullorum status of a flock should be exercised only after conclusive evidence of infection has been established.

## THE STAINED-ANTIGEN, RAPID WHOLE-BLOOD TEST

The stained-antigen, rapid whole-blood test was first developed by Schaffer, MacDonald, Hall, and Bunyea (1931), and Coburn and Stafseth (1931). At the present time, the antigen for this method is produced under federal license from the Secretary of Agriculture, in accordance with specific directions.

For a detailed description of the official procedure, reference should be made to the revised report on The National Poultry Improvement Plan (Anon., 1946). Briefly, the method may be described as follows: A wire loop, three-sixteenths of an inch in diameter made on the end of a 2½-inch length, noncorrosive wire (Brown and Sharpe gauge No. 20), is used to

measure the blood. One end of the wire is inserted into a cork stopper which serves as a handle. A loopful of blood is taken up from the punctured wing vein and contains approximately .02 cc. when the blood appears to bulge out. The loopful of blood is mixed with the stained antigen which has been placed on a glass plate marked off in inch squares. The antigen is measured with a medicine dropper whose tip is constructed to deliver .05 cc. when held in a vertical position. An antigen-blood dilution of two to one or three to one has been reported to give the most satisfactory results. The loopful of blood is mixed with the antigen, and the mixture is spread out about an inch in diameter. The loop is washed in clean water and dried with cheese cloth or blotting paper. The glass plate is tilted up and down several times to aid in the mixing of the blood and antigen and apparently has some influence on the speed of the agglutination. Reactions may occur within a few seconds up to 2 minutes. Delayed reactions should be regarded as nonspecific. A positive reaction consists of the clumping of the violet stained cells floating in clear fluid (Fig. 8.21).



Fig. 8.21. Stained antigen, whole-blood test. Left— negative reaction. Right—positive reaction.

The rapidity of the reaction and the size of the clumps are influenced by the agglutinating power of the blood. Partial reactions should be regarded as suspicious and treated in the same manner as those observed in the other testing methods. Sometimes a very fine granulation appears which should be considered negative. Very infrequently agglutination of the red blood cells occurs and should not be confused with the clumping of the stained bacterial cells. The fine marginal flocculation which may be observed before drying of the mixture is to be considered negative. A negative reaction is one in which the mixture remains homogeneous for at least 2 minutes. Recently the Federal Bureau of Animal Industry (MacDonald, 1947) has developed an antigen which was designated as the K antigen. The cultures for the antigen are grown on a special medium. It is reported that the new medium produces a higher yield of organisms which possess a greater antigenic specificity than in other stained antigens recommended previously.

Only those reactions which appear within 1 minute after the mixing of the blood and antigen should be considered definitely positive, while reactions delayed for 2 minutes should be considered suspicious.

In order to approach uniformity of results, the testing plate should be well lighted at all times, and the temperature should remain at a constant
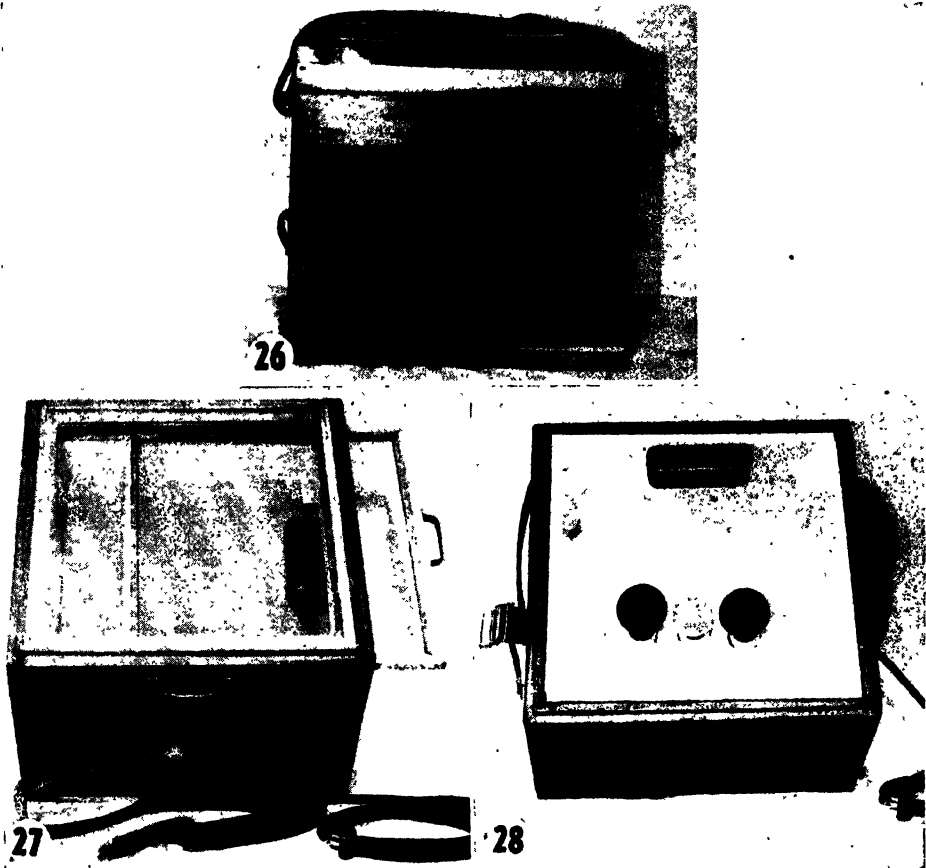


Fig. 8.22. 26—an electrically heated and lighted apparatus used for the stained antigen, whole-blood test. 27—same apparatus with cover removed. Test plate slides into a removable frame which is covered with glass. 28—same apparatus exposing the two black heat bulbs and a white light bulb.

level. A temperature of 75–85° F. is considered satisfactory. The test plate should be free of dust and so constructed that it can be tilted with ease (Fig. 8.22). The tested birds can be retained in either special holding equipment or crates and released as rapidly as the results of the test become known (Fig. 8.23). All birds in the tested flock should be officially leg-banded. The accuracy of the results is greatly influenced by the competency of the testing agent and his thoroughness and care in conducting the test.

### THE RAPID SERUM TEST

The rapid serum test for the detection of pullorum disease carriers was developed by Runnells, Coon, Farley, and Thorp (1927). The blood samples may be collected in a manner similar to that described for the tube test. The antigen employed consisted of a single strain of *S. pullorum* suspended in 12 per cent sodium chloride solution containing 0.5 per cent of
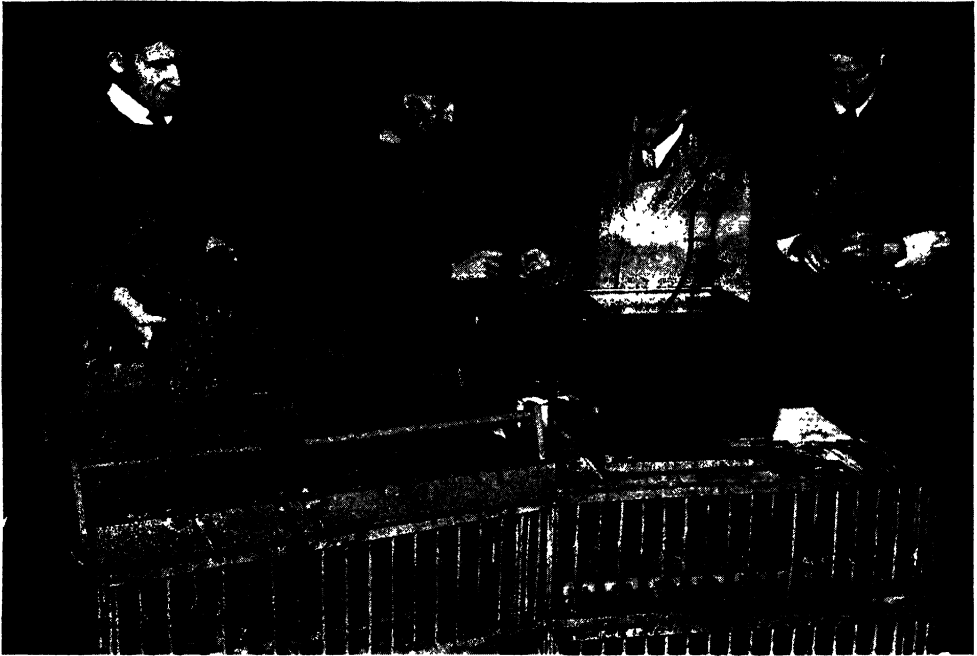


FIG. 8.23. Conducting the stained antigen, whole-blood test in the poultry house.

phenol. The turbidity was adjusted to 50 times greater than tube 0.75 of McFarland's nephelometer.

A box with a glass top ruled off in inch squares and improvised with lighting and heating facilities was used for testing. Two serum-antigen dilutions corresponding to the 1:50 and 1:100 dilutions for the tube test were employed. The amounts of serum used were 0.02 cc. and 0.01 cc. to which was added 0.02 cc. of antigen. The serum and antigen were mixed thoroughly with a toothpick. Positive reactions may occur quickly, but delayed reactions may require several minutes (Fig. 8.24). Gradations of reactions occur in this method as in other methods. Considerable experience is necessary for proper interpretation. This method should be used only in competent hands if the results are to be regarded as official. The results of the tests and the numbers of spoiled, broken, and missing samples should be reported directly to the flock owner or the agency in charge of the field work.

### INTRADERMAL TEST

Ward and Gallagher (1917) reported the application of an intracutaneous or intradermal test based on allergic reactions for the detection of carriers of pullorum disease. Later this method was extensively investigated by others (Bushnell and Brandly, 1929; Edwards and Hull, 1929; Michael and Beach, 1929; Rettger, McAlpine, and Warner, 1930). Cellular suspensions or extracts of *S. pullorum* were treated in various ways and then injected intracutaneously into the wattle of the bird. The production of a swelling was considered a positive reaction. The term "pullorin test" also has been applied to this method of detecting carriers. However, the investigational findings and practical results revealed that the method is not sufficiently reliable for practical use in the control and eradication of the disease.

Other tests (Edwards and Hull, 1929) such as the precipitin test and the complement-fixation method were found either unreliable or impracticable in control and eradication programs.

In the control and eradication of the disease, the actual detection of infected birds through testing is only an integral part of a large program. A testing program which does not consider all the ramifica-



FIG. 8.24. The rapid serum agglutination test. Left—negative reaction. Right—positive reaction.

tions of pullorum disease is doomed to meet failure. Indiscriminate testing for the purpose of advertising or promoting the sale of stock should be prohibited. An adequate testing program should give consideration to the following eradication and preventive measures:

1. All birds over five months of age should be tested annually in order to determine the true status of a flock. When infection exists in the flock, partial flock testing is not as effective in eliminating the disease from the premises as is 100 per cent testing.

On commercial farms where large units of birds are maintained for egg production in addition to units held for breeding purposes, the testing of only the breeding stock might suffice provided proper facilities exist for the segregation of the two groups and adequate precautionary measures are carried out against direct or indirect contact between the two groups. One

should not consider a plant having both untested and tested birds as safe as a plant with only tested birds. A keen and careful buyer of stock will always carefully investigate this point and buy only from a flock whose status is without doubt.

Intermittent testing, that is, testing one year and not the next, or on alternate years, is a procedure which is not effective in establishing or maintaining a free flock. Those engaged in an official testing program should adopt the policy of minimizing such a practice.

TABLE 1

RETESTING DATA OF TEN INFECTED FLOCKS

| Flock Number | | First Test | Second Test | Third Test | Fourth Test | Fifth Test | Results of Subsequent Season |
|---|---|---|---|---|---|---|---|
| 1....... | No. of birds tested... | 189 | 152 | 48 | ........ | ........ | 467 |
| | Percentage reactors.. | 1.59 | 0.00 | 0.00 | ........ | ........ | 0.00 |
| 2....... | No. of birds tested... | 369 | 256 | 218 | ........ | ........ | 232 |
| | Percentage reactors.. | 0.54 | 0.39 | 0.00 | ........ | ........ | 0.00 |
| 3....... | No. of birds tested... | 125 | 98 | 91 | ........ | ........ | 201 |
| | Percentage reactors.. | 20.00 | 4.08 | 0.00 | ........ | ........ | 0.00 |
| 4....... | No. of birds tested... | 243 | 262 | 223 | 179 | ........ | 199 |
| | Percentage reactors.. | 11.11 | 1.15 | 0.00 | 0.00 | ........ | 0.00 |
| 5....... | No. of birds tested... | 464 | 444 | 433 | 397 | ........ | 1,087 |
| | Percentage reactors.. | 2.37 | 0.45 | 0.00 | 0.00 | ........ | 0.00 |
| 6....... | No. of birds tested... | 1,765 | 1,559 | 1,508 | 1,108 | 767 | 1,796 |
| | Percentage reactors.. | 3.17 | 0.13 | 0.00 | 0.00 | 0.00 | 0.00 |
| 7....... | No. of birds tested... | 2,079 | 1,929 | 1,811 | 1,648 | 1,337 | 2,132 |
| | Percentage reactors.. | 3.17 | 1.09 | 0.00 | 0.00 | 0.00 | 0.00 |
| 8....... | No. of birds tested... | 704 | 691 | 610 | 422 | ........ | 693 |
| | Percentage reactors.. | 8.24 | 8.83 | 0.16 | 0.00 | ........ | 0.00 |
| 9....... | No. of birds tested... | 2,722 | 2,413 | 2,284 | 1,929 | ........ | 3,707 |
| | Percentage reactors.. | 1.80 | 0.54 | 0.48 | 0.00 | ........ | 0.00 |
| 10....... | No. of birds tested... | 640 | 440 | 399 | 352 | 339 | 747 |
| | Percentage reactors.. | 27.34 | 4.32 | 1.00 | 0.00 | 0.00 | 0.00 |

2. Flocks revealing infection should be retested within four or six weeks until a negative report is obtained provided the value of the birds justified the expenditure. In the majority of cases, infection can be eliminated from a flock through short interval testing. Two or three retests in many instances are sufficient to detect all the infected birds in a flock (Table 1). Occasionally, infection may be very persistent, so that its elimination may not yield to a testing program.

A retesting program for an infected flock or flocks should be complete to the extent that all infected birds have been detected and removed. Permit-

ting one or a few infected birds to remain in a flock after a partial retesting program has been applied may lead to the propagation of the infection in the progeny of that flock which will necessitate a program of multiple testing from year to year to combat the disease. The objective should be to eliminate all of the infected birds from the breeding flock and reduce the cost of testing to one annual test in order to determine the status of the flock. In areas where the majority of flocks are free of the disease, the need for multiple retesting has been eliminated, which consequently represents a great economy to the poultry industry.

Infected flocks of inferior breeding or revealing heavy infection should not be considered for retesting. Replacements from known pullorum clean sources will prove more effective and less expensive than intensive retesting for the establishment of a free flock. However, the pullorum disease-free stock selected for replacements should be protected against reinfection, which is not always fully appreciated. In some states the policy of replacement of infected flocks with stock from free sources has contributed more to the eradication of the disease and at less cost than could have been accomplished through retesting. In areas where the majority of flocks still harbor the infection, it would behoove the official state agencies, commercial hatchery-men, and flock owners to carefully consider ways and means whereby similar progress can be made in their respective localities. Once the goal of pullorum freedom has been attained, a retrogression to pullorum infected flocks will not be tolerated.

3. Every reactor, regardless of its value, should be removed from the premises and sold for slaughter immediately upon the completion of the test. Reactors should not be retained or sold for egg production because they would serve as sources for the spread of the disease.

4. The poultry houses, runs, and equipment should be thoroughly cleaned and disinfected immediately after removal of reactors. An empty pen in each house will facilitate cleaning and disinfection during the winter months. Disinfectants approved by federal or state agencies are recommended. Occasionally, circumstances are such that an empty pen is not available, but this should not prohibit thorough cleaning of the pens which is highly essential in the eradication of the disease. Van Roekel, Bullis, Flint, and Clarke (1941) have observed that S. *pullorum* may remain viable on a dry cloth maintained at room temperature for at least seven years and eight months. Allen and Jacob (1930) found the organism to persist in a virulent condition for at least fourteen months in samples of contaminated soil and deduced from this that the infection could exist on a premise from one season to the next. Kerr (1930) observed that S. *pullorum* remained viable in fecal emulsions for more than three months. It appears that soil in runs or yards inhabited by infected flocks should be considered unsafe for pullorum-free

stock. Placing such stock on new ground is a highly effective means against contracting the disease from contaminated soil. Frequent plowing and liming of contaminated soil will aid in the destruction of the organism.

5. Offal from all birds dressed for market or home consumption, as well as dead birds that are unfit for consumption, should be burned. Feeding of garbage should be avoided.

6. Eggs should not be saved for hatching until after a flock has been tested and found to be free of the disease. As long as infected birds are detected in a flock, one may anticipate losses from the disease among the progeny.

7. Fresh and infertile eggs from unknown or infected sources should not be fed to chickens or exposed to animals such as crows, sparrows, rats, and
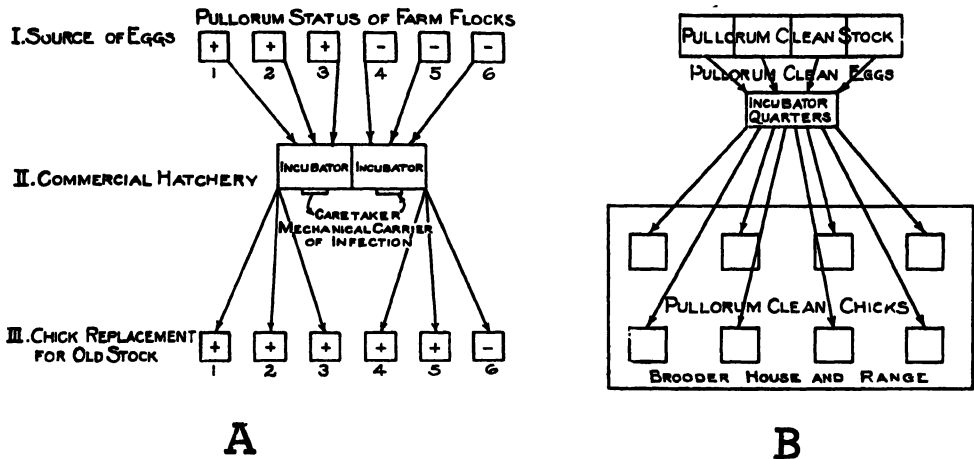


FIG. 8.25. A—an ineffective control and eradication plan for pullorum disease. B—a successful eradication plan for pullorum disease.

skunks that may carry or spread the infection. Investigations (Van Roekel, Bullis, Flint, and Clarke, 1932) definitely reveal that infection may be contracted from such sources.

8. Poultrymen should not custom hatch for untested or infected flocks (including fowl other than chickens) in view of the fact that the infection can be transmitted through the incubator. This likewise applies to commercial hatcheries. In some sections commercial hatcheries select hatching eggs largely from infected flocks (Fig. 8.25). Facilities for separately maintaining eggs from infected and noninfected sources often do not exist in commercial hatcheries. Little progress in the eradication of disease can be expected where such conditions prevail. Commercial hatcheries might well provide for hatching facilities entirely separate from the main hatchery for the purpose of producing chicks for replacements of their supply flocks. In a

plan of this nature hatching eggs should be selected from pullorum-free flocks. Such a program has been in effect in certain localities and has proven successful in the eradication of the disease from flocks. The breeder-hatcher plan (the breeder conducts the hatching) has been highly successful in the establishment and maintenance of pullorum-free flocks (Fig. 8.25). In states where this plan is operating, 90–95 per cent of the total birds tested are in pullorum-free flocks. Very few "breaks" occur from year to year. The amount of infection in these flocks as a rule is slight, and its source in most instances may be attributed to inadequate prevention.

9. The purchase of stock in the form of adults, chicks, and eggs should be from known pullorum disease-free flocks. Official state agencies or their published lists of pullorum-free flocks should be consulted as a guide in the purchase of stock. Purchases should not be made on advertisements or sales literature alone, because of the lack of information or misleading statements.

TABLE 2

TWENTY-SEVEN-YEAR PULLORUM DISEASE TESTING SUMMARY FOR MASSACHUSETTS

| Season | Flocks | Birds | Total Tests | Positive Tests Percentage | Non-reacting Flocks | Birds in Non-reacting Flocks | |
| --- | --- | --- | --- | --- | --- | --- | --- |
| | | | | | | Number | Percentage |
| 1920–21.... | 108 | 24,718 | 24,718 | 12.50 | 25 | 2,414 | 9.77 |
| 1921–22.... | 110 | 29,875 | 29,875 | 12.65 | 27 | 4,032 | 13.50 |
| 1922–23.... | 121 | 33,602 | 33,602 | 7.60 | 29 | 5,400 | 16.07 |
| 1923–24.... | 139 | 59,635 | 59,635 | 6.53 | 38 | 11,082 | 18.58 |
| 1924–25.... | 156 | 66,503 | 66,503 | 2.94 | 79 | 25,390 | 38.18 |
| 1925–26.... | 201 | 67,919 | 67,919 | 2.31 | 124 | 33,615 | 49 49 |
| 1926–27.... | 249 | 127,327 | 127,327 | 4 03 | 114 | 40,269 | 31.63 |
| 1927–28 ... | 321 | 190,658 | 232,091 | 6.52* | 138 | 80,829 | 42.39 |
| 1928–29.... | 413 | 254,512 | 304,092 | 4.25* | 228 | 153,334 | 60.25 |
| 1929–30.... | 460 | 331,314 | 386,098 | 2.17 | 309 | 203,038 | 66.97 |
| 1930–31.... | 447 | 356,810 | 402,983 | 1.47 | 328 | 267,229 | 74.89 |
| 1931–32.... | 455 | 377,191 | 420,861 | 0.90 | 355 | 298,534 | 79.15 |
| 1932–33.... | 335 | 296,093 | 300,714 | 0.47 | 276 | 238,074 | 80.41 |
| 1933–34.... | 262 | 263,241 | 284,848 | 0.53 | 229 | 212,782 | 80.83 |
| 1934–35.... | 244 | 281,124 | 301,887 | 0.39 | 213 | 251,778 | 89.56 |
| 1935–36.... | 252 | 329,659 | 344,081 | 0.30 | 230 | 315,215 | 95.95 |
| 1936–37.... | 307 | 448,519 | 461,762 | 0.37 | 281 | 424,431 | 94.63 |
| 1937–38.... | 308 | 480,227 | 497,769 | 0.17 | 286 | 457,466 | 95.26 |
| 1938–39.... | 355 | 571,065 | 615,205 | 0.34 | 327 | 469,134 | 82.15 |
| 1939–40.... | 346 | 573,000 | 673,222 | 0.51 | 332 | 497,356 | 86.80 |
| 1940–41.... | 309 | 527,328 | 538,589 | 0.09 | 299 | 492,475 | 93.39 |
| 1941–42.... | 366 | 653,080 | 662,715 | 0.27 | 350 | 591,628 | 90.59 |
| 1942–43.... | 332 | 637,666 | 649,137 | 0.48 | 317 | 600,607 | 94.19 |
| 1943–44.... | 413 | 762,066 | 791,596 | 0.11 | 386 | 721,229 | 94.64 |
| 1944–45.... | 458 | 836,481 | 943,987 | 0.12 | 431 | 792,551 | 94.75 |
| 1945–46.... | 538 | 1,125,737 | 1,225,594 | 0.12 | 513 | 1,085,726 | 96.45 |
| 1946–47.... | 562 | 1,156,147 | 1,238;983 | 0.13 | 534 | 1,112,043 | 96.19 |

* Based on total birds tested: 1927–28, 190,658 birds; 1928–29, 254,512 birds.

The breeder-hatcher, commercial hatcherymen, and buying public have a common objective, and that is to produce healthy profitable poultry.

10. Birds removed from the premises to egg-laying contests and exhibitions should be held in quarantine and determined free of the disease before they are readmitted into the flock. In several instances the source of pullorum infection was traceable to contests and shows. Birds returned to the premises should be tested immediately upon their arrival and retested after 30 days' quarantine. The safest procedure is not to return such birds to the

TABLE 3

PULLORUM DISEASE TESTING SUMMARY OF THIRTEEN STATES COVERING A TWENTY YEAR PERIOD*

| Item | 1927–28 | 1937–38 | 1946–47 |
|---|---|---|---|
| Number of tests............................ | 735,851 | 3,129,344 | 9,694,772 |
| Percentage of positive tests...................... | 3.2 | 1.7˙ | 0.52 |
| Number of negative flocks (100 per cent tested)..... | 372 | 2,564 | 7,693 |
| Number of breaks in negative flocks | | | |
| (100 per cent tested)...................... | 43 | 275 | 788 |
| Number of pullorum clean flocks. ............. . | 201 † | 1,222 | 4,279 |
| Number of birds in pullorum clean flocks.......... | 112,605 | 1,107,118 | 4,503,426 |

* Includes Connecticut, Delaware, Maine, Maryland, Massachusetts, New Hampshire, New Jersey, New York, Pennsylvania, Rhode Island, Virginia, Vermont, and West Virginia.

† Massachusetts did not recognize an official grade, which requires a flock to pass two consecutive negative tests not less than 6 months nor more than a year apart.

breeding flock, to avoid introduction of other diseases as well as pullorum disease.

11. Fowl other than chickens should be considered as a possible source of infection. The testing of such fowl may aid in determining their pullorum status. The eggs from chickens and from fowl other than chickens should not be hatched simultaneously in the same incubator. This precaution will avoid the spread of paratyphoid infections as well as pullorum disease among the different species of fowl.

12. Used feed bags and other equipment that may have been exposed to or contaminated with infective material should not be used unless properly cleaned and disinfected. Dunlap (1931) reported transmission of the disease to chicks which were fed mash from an artificially contaminated bag.

*Progress in Control and Eradication.* Considerable progress in the actual control and eradication of the disease has been made during the last decade. This is true not only in the United States but also in other countries. In some sections of the United States, especially the northeastern states, exceedingly great progress in the establishment of pullorum-free flocks has been made (Tables 2 and 3). With the adoption of the National Poultry Improvement Plan (Simms, 1946), a growing interest in pullorum disease has become manifest, and many infected birds are being detected and removed from

breeding flocks under an official program. Forty-seven states are officially cooperating with the national government in the pullorum phase of the plan. The number of birds tested under the plan increased from 2,058,782 in 1936 to 22,294,650 in 1946. The average incidence of reactors among flocks was below 2 per cent. In some sections of this country marked progress has been made toward establishing flocks free of pullorum disease and recognizing only flocks with no pullorum tolerance. While nationwide progress has been made in reducing chick losses from pullorum disease, greater emphasis should be given to participation in the grades which tolerate no reactors.

Agencies in charge of the control and eradication of pullorum disease should enlist the cooperative efforts of veterinarians, flock owners, hatchery-men, poultry organizations, and county extension services for an educational program to promote further interest in the establishment and maintenance of pullorum-free flocks.

## REFERENCES

Allen, P. W., and Jacob, M.: 1930. Sodium acid sulphate as a disinfectant against *Salmonella pullorum* in poultry-yard soils. Tenn. Agr. Exper. Sta., Bul. 143.

Anonymous: 1930. Eastern States Conference of Laboratory Workers in Pullorum Disease Control. Jour. Am. Vet. Med. Assn. 77:259.

Anonymous: 1933. Report of the conference of official research workers in animal diseases of North America on standard methods of pullorum disease in barnyard fowl. Jour. Am. Vet. Med. Assn. 82:487.

Anonymous: 1941. The National Poultry Improvement Plan. U. S. D. A. Misc. Pub. 300:28.

Anonymous: 1946. The National Poultry Improvement Plan. U. S. D. A. Misc. Pub. 300:28.

Asmundson, V. S., and Biely, J.: 1930. Effect of pullorum disease on distribution of first year egg production. Scient. Agr. 10:497.

Barboni, E.: 1937. Ricerche sul primo focolaio di pullorosi nei tacchini riscontrato in Italia. Clin. Vet. 60:597.

Beach, J. R., and Freeborn, S. B.: 1927. Diseases and parasites of poultry in California. Calif. Agr. Ext. Ser., Cir. 8, 5th edition:110.

Beaudette, F. R.: 1938. Localized pullorum infection in the ovary of a duck. Jour. Am. Vet. Med. Assn. 92:100.

——, Bushnell, L. D., and Payne, L. F.: 1923. Relation of *Bacterium pullorum* to hatchability of eggs. Jour. Infect. Dis. 32:331.

Benedict, R. G., McCoy, E., and Wisnicky, W.: 1941. Salmonella types in silver foxes. Jour. Infect. Dis. 69:167.

Bottorff, C. A., and Kiser, J. S.: 1946. The use of sulfonamides in the control of pullorum disease. Poultry Sci. 25:397.

Brunett, E. L.: 1930. Pullorum disease in the mature turkey. Poultry Sci. 9:356.

Bunyea, H.: 1939. An outbreak of pullorum disease in young guinea fowl. Jour. Am. Vet. Med. Assn. 94:233.

—— and Hall, W. J.: 1929. Some observations on the pathology of bacillary white diarrhea in baby chicks. Jour. Am. Vet. Med. Assn. 75:581.

Burton, W. H.: 1946. Control of pullorum disease transmission in hatcheries by formaldehyde fumigation. Proc. 18th Ann. Conf. Lab. Workers in Pullorum Disease Control, p. 1.

Bushnell, L. D., and Brandly, C. A.: 1929. Some experiments on the control of bacillary white diarrhea. Jour. Am. Vet. Med. Assn. 74:444.

——, Hinshaw, W. R., and Payne, L. F.: 1926. Bacillary white diarrhea in fowl. Kan. Agr. Exper. Sta., Tech. Bul. 21.

—— and Payne, L. F.: 1931. Dissemination of pullorum disease in the incubator. Kan. Agr. Exper. Sta., Tech. Bul. 29.

——, Payne, L. F., and Coon, C. J.: 1929. Fumigation of force-draft incubators. Jour. Am. Vet. Med. Assn. 75:611.

—— and Porter, J. J.: 1915. A study of methods for the isolation of *Salmonella pullorum*. Poultry Sci. 24:212.

Coburn, D. R., and Stafseth, H. J.: 1931. A field test for pullorum disease. Jour. Am. Vet. Med. Assn. 79:241.

Dalling, T., Mason, J. H., and Gordon, W. S.: 1928. Bacillary white diarrhea (B.W.D.) : *B. pullorum* isolated from sparrows. Vet. Record 8:329.

———, Mason, J. H., and Gordon, W. S.: 1929. Bacillary white diarrhea (B.W.D.) : *B. pullorum* isolated from a turkey poult in England. Vet. Record 9:902.

Dearstyne, R. S., Kaupp, B. F., and Wilfong, H. S.: 1929. Study of bacillary white diarrhea (pullorum disease). Agr. Exper. Sta. of N. C. State Coll. of Agr. and Eng., and N. C. Dept. of Agr., Tech. Bul. 36:53.

DeVolt, H M., Quigley, C. D., and Byerly, T. C.: 1941. Studies of resistance to pullorum disease in chickens. Poultry Sci. 20:339.

Doyle, L. P., and Mathews, F. P.: 1928. The pathology of bacillary white diarrhea in chicks. Purdue Univ. Agr. Exper. Sta., Bul. 323.

Doyle, T. M.: 1925. Bacillary white diarrhea of chicks. Jour. Comp. Path. and Therap. 38:266.

Dunlap, G. L.: 1931. Ann. Rep., Mass. Agr. Exper. Sta., Bul. 271:280–81.

E. A. H.: 1905. White diarrhea in brooder chicks. Farm Poultry 16, No. 9:256.

Edwards, P. R.: 1928. The fermentation of maltose by *Bacterium pullorum*. Jour. Bact. 15:235.

———: 1945. An outbreak of *Salmonella pullorum* infection in canaries. Jour. Am. Vet. Med. Assn. 107:245.

——— and Bruner, D. W.: 1943. The occurrence and distribution of Salmonella types in the United States. Jour. Infect. Dis. 72:58.

——— and Bruner, D. W.: 1946. Form variation in *Salmonella pullorum* and its relation to X strains. Cornell Vet. 36:318.

——— and Hull, F. E.: 1929. Bacillary white diarrhea and related diseases of chickens. Kentucky Agr. Exper. Sta., Bul. 296:237–80.

Emmel, M. W.: 1936. Pullorum disease in captive quail. Jour. Am. Vet. Med. Assn. 89:716.

Felsenfeld, O., and Young, V. M.: 1944. The occurrence of members of the genus Salmonella in inhabitants of state hospitals of the greater Chicago area. Jour. Lab. and Clin. Med. 29:375.

Gage, G. E., Paige, B. H., and Hyland, H. W.: 1914. On the diagnosis of infection with *Bacterium pullorum* in the domestic fowl. Mass. Agr. Exper. Sta., Bul. 148.

Garrard, E. H., Burton, W. H., and Carpenter, J. A.: 1947. Non-pullorum reactions. Proc. 19th Ann. Conf. Lab. Workers in Pullorum Disease Control. p. 1.

Gifford, E. G.: 1905. White diarrhea in incubator chicks. Farm Poultry 16, No. 10:269.

Graham, R.: 1941. Hatchery sanitation and incubator hygiene. Proc. Conf. Nat. Poultry Improv. Plan, Bur. of An. Ind., U.S.D.A., 106–15.

Graham, W. R.: 1904. White diarrhea in brooder chicks. Farm Poultry 15, No. 11:252.

Gwatkin, R.: 1927. Some experiments on the disinfection of eggs and incubators. Ont. Vet. Coll. Rep. 1926. 58–65.

———: 1946. Resumé of studies relating to Younie strains of *Salmonella pullorum*. Proc. 18th Ann. Conf. Lab. Workers in Pullorum Disease Control, p. 1.

——— and Glover, J. S.: 1930. Isolation of *S. pullorum* from the nasal passages of two fowl. Ont. Vet. Coll. Rep. 1929:61.

——— and Mitchell, C. A.: 1944. Transmission of *Salmonella pullorum* by flies. Canad. Jour. Pub. Health 35:281.

Hanks, J. H., and Rettger, L. F.: 1932. Bacterial endotoxin. Search for a specific intracellular toxin in *S. pullorum*. Jour. Immunol. 22:283.

Hendrickson, J. M.: 1927. The differentiation of *Bacterium pullorum* (Rettger) and *Bacterium sanguinarium* (Moore) . Jour. Am. Vet. Med. Assn. 70:629.

——— and Hilbert, K. F.: 1930. Report of the Poultry Disease Laboratory at Farmingdale, Long Island. Ann. Rep. N. Y. State Vet. Coll. 1928–29:49–53.

——— and Hilbert, K. F.: 1931. Report of the Poultry Disease Laboratory at Farmingdale, Long Island. Ann. Rep. N. Y. State Vet. Coll. 1929–30:51–55.

Hewitt, E. A.: 1928. Bacillary white diarrhea in baby turkeys. Cornell Vet. 18:272.

Hinshaw, W. R.: 1937. Diseases of turkeys. Univ. of Calif. Agr. Exper. Sta., Bul. 613.

———: 1939. Pullorum disease in turkeys. Proc. Conf. Nat. Poultry Improv. Plan, Bur. An. Ind., An. Hus. Div., U.S.D.A., 98–104.

———: 1941. Cysteine and related compounds for differentiating members of the genus Salmonella. Hilgardia 13, No. 11:583–621.

——— and Hoffman, H. A.: 1937. Pullorum disease in ducklings. Poultry Sci. 16:189.

———, Upp, C. W., and Moore, J. M.: 1926. Studies in transmission of bacillary white diarrhea in incubators. Jour. Am. Vet. Med. Assn. 68:631.

Hudson, C. B., and Beaudette, F. R.: 1929. The isolation of *Bacterium pullorum* from a European bullfinch (*Pyrrhula europa*) . Jour. Am. Vet. Med. Assn. 74:929.

Hutt, F. B., and Scholes, J. C.: 1941. Genetics of the fowl. XIII. Breed differences in susceptibility to *Salmonella pullorum*. Poultry Sci. 20:342.

Hutyra, F., Marek, J., and Manninger, R.: 1938. Special Pathology and Therapeutics of the Diseases of Domestic Animals. Vol. I, 4th English edition. Alexander Eger, Chicago. 212–24.

Insko, W. M., Steele, D. G., and Hinton, C. M.: 1941. Effect of formaldehyde fumigation on mortality of chick embryos. Kentucky Agr. Exper. Sta., Bul. 416.

Jones, F. S.: 1911. Fatal septicemia or bacillary white diarrhea in young chickens. Ann. Rep. N. Y. State Vet. Coll. 1909–10:111–29.

————: 1913a. An outbreak of an acute disease in adult fowls due to *Bacterium pullorum*. Jour. Med. Res. 27:471.

————: 1913b. The value of the macroscopic agglutination test in detecting fowls that are harboring *Bacterium pullorum*. Jour. Med. Res. 27:481.

Jungherr, E.: 1935. Diseases of brooder chicks. Storrs Agr. Exper. Sta., Bul. 202:56.

Kaupp, B. F.: 1917. Poultry Diseases. 2nd edition, Revised and enlarged. Am. Vet. Pub. Co., Chicago. 119–24.

Kerr, W. R.: 1930. Selective media for the cultivation of *Bacillus pullorum* and *Bacillus sanguinarium*. Jour. Comp. Path. and Therap. 43:77.

Lerche, M.: 1929. Ueber das Vorkommen der bakteriellen (weissen) Kückenruhr bei jungen Enten. Tierärztl. Rundschau. 35:169.

MacDonald, A. D.: 1947. K antigen for the detection of pullorum disease in poultry. Proc. 19th Ann. Conf. Lab. Workers in Pullorum Disease Control, p. 1.

Mallmann, W. L.: 1929. *Salmonella pullorum* in the intestinal contents of baby chicks. Jour. Infect. Dis. 44:16.

————: 1931a. Studies on bacteriophage in relation to Salmonella and pullorum disease. Mich. Agr. Exper. Sta., Tech. Bul. 109.

————: 1931b. Use of organic acids for the differentiation of *Salmonella pullorum* and *Salmonella gallinarum*. Proc. Soc. Exper. Biol. and Med. 28:501.

————: 1932. The dissociation of *Salmonella pullorum* and related species. Mich. Agr. Exper. Sta., Tech. Bul. 122.

———— and Snyder, D.: 1929. Differential medium for *Salmonella pullorum, Salmonella gallinarum, Pasteurella avicida*, and *Escherichia coli*. Jour. Infect. Dis. 44:13.

Mathews, F. P.: 1927. Factors influencing the control of bacillary white diarrhea. Jour. Am. Vet. Med. Assn. 71:585.

Michael, S. T., and Beach, J. R.: 1929. An experimental study of tests for the detection of carriers of *Bacterium pullorum*. Hilgardia 4, No. 8:185–200.

Miessner, H.: 1931. Bacillary white diarrhea—Fowl typhoid. Proc. Fourth World's Poultry Cong. 1930 (London), No. 64:428.

Mitchell, R. B., Garlock, F. C., and Broh-Kahn, R. H.: 1946. An outbreak of gastro-enteritis presumably caused by *Salmonella pullorum*. Jour. Infect. Dis. 79:57.

Moore, J. M., Mallmann, W. L., and Arnold, L. R.: 1934. Studies on pullorum disease. I. The influence of different temperatures in brooding. Jour. Am. Vet. Med. Assn. 84:526.

Mulsow, F. W.: 1919. The differentiation and distribution of the paratyphoid-enteritidis group. VI. Avian paratyphoid bacilli: a comparative study of *Bacterium pullorum* and *Bacterium sanguinarium*. Jour. Infect. Dis. 25:135.

Olney, J. F.: 1928. *Salmonella pullorum* infection in rabbits. Jour. Am. Vet. Med. Assn. 73:631.

Pacheco, G., and Rodrigues, C.: 1936. O grupo pullorum-gallinarum em provas bacteriologicas comparativas. Inst. Oswaldo Cruz 31:591–705.

Plastridge, W. N., and Rettger, L. F.: 1930. An epidemic disease of domestic fowl caused by a hitherto undescribed organism of the *Salmonella pullorum* type. Jour. Infect. Dis. 47:334.

———— and Rettger, L. F.: 1932. Variants of *Salmonella pullorum*. Jour. Infect. Dis. 50:146.

Reis, J., and Nobrega, P.: 1936. Tratado de doencas das aves. Edicao do Instituto Biologico, São Paulo, Brazil:109.

Rettger, L. F.: 1900. Septicemia among young chickens. N. Y. Med. Jour. 71:803.

————: 1901. Septicemia in young chickens. N. Y. Med. Jour. 73:267.

————: 1909. Further studies on fatal septicemia in young chickens, or "white diarrhea." Jour. Med. Res. 21:115.

————: 1916. Occurrence and significance of *Bacterium pullorum* in eggs. Jour. Am. Assn. Instr. and Invest. Poultry Husb. 2:62.

————, Hull, T. G., and Sturges, W. S.: 1916. Feeding experiments with *Bacterium pullorum*. The toxicity of infected eggs. Jour. Exper. Med. 23:475.

————, Kirkpatrick, W. F., and Jones, R. E.: 1914. Bacillary white diarrhea of young chicks. (Fourth report) Storrs Agr. Exper. Sta., Bul. 77:259–309.

————, Kirkpatrick, W. F., and Jones, R. E.: 1915. Bacillary white diarrhea of young chicks: its eradication by the elimination of infected breeding stock. (Fifth report) Storrs Agr. Exper. Sta., Bul. 85:149–67.

————, McAlpine, J. G., and Warner, D. E.: 1930. A comparative study of the routine macroscopic agglutination and the intracutaneous (wattle) tests for *Bacterium pullorum* infection in poultry breeding stock. Jour. Am. Vet. Med. Assn. 77:47.

———— and Plastridge, W. N.: 1932. Pullorum disease of domestic fowl. Monograph. Storrs Agr. Exper. Sta., Bul. 178:103–92.

———— and Stoneburn, F. H.: 1909. Bacillary white diarrhea of young chicks. Storrs Agr. Exper. Sta., Bul. 60:29–57.

———— and Stoneburn, F. H.: 1911. Bacillary white diarrhea of young chicks. (Second report.) Storrs Agr. Exper. Sta., Bul. 68:275–301.

Roberts, E., and Card, L. E.: 1935. Inheritance of resistance to bacterial infection in animals. A genetic study of pullorum disease. Ill. Agr. Exper. Sta., Bul. 419:467–93.

Roberts, E., Severens, J. M., and Card, L. E.: 1939a. Effect of environment on the expression of resistance and susceptibility to disease in the domestic fowl. Proc. of the Seventh World's Poultry Cong., Cleveland. 431.

———, Severens, J. M., and Card, L. E.: 1939b. Nature of the hereditary factors for resistance and susceptibility to pullorum disease in the domestic fowl. Proc. of the Seventh World's Poultry Cong., Cleveland. 52.

Runnells, R. A.: 1929. Bacillary white diarrhea. Pullorum infection of the domestic fowl. Va. Agr. Exper. Sta., Bul. 265:27.

———, Coon, C. J., Farley, H., and Thorp, F.: 1927. An application of the rapid-method agglutination test to the diagnosis of bacillary white diarrhea infection. Jour. Am. Vet. Med. Assn. 70:660.

——— and Van Roekel, H.: 1927a. The occurrence of white diarrhea infection in eggs laid by hens reacting to the agglutination test. Poultry Sci. 6:141.

——— and Van Roekel, H.: 1927b. Further observations on the occurrence of white diarrhea infection in eggs laid by hens reacting to the agglutination test. Poultry Sci. 6:229.

Salmonella Subcommittee of the Nomenclature Committee of the International Society for Microbiology: 1934. The genus *Salmonella*, Lignières, 1900. Jour. of Hyg. 34:333.

Schaffer, J. M., MacDonald, A. D., Hall, W. J., and Bunyea, H.: 1931. A stained antigen for the rapid whole blood test for pullorum disease. Jour. Am. Vet. Med. Assn. 79:236.

Scholes, J. C.: 1942. Experiments with X-rays on the roles of lymphocytes and body temperatures in the resistance of chicks to *Salmonella pullorum*. Poultry Sci. 21:561.

——— and Hutt, F. B.: 1942. The relationship between body temperature and genetic resistance to *Salmonella pullorum* in the fowl. Cornell Univ. Agr. Exper. Sta. Memoir 244:1–35.

Severens, J. M., Roberts, E., and Card, L. E.: 1945. The effect of sulfonamides in reducing mortality from pullorum disease in the domestic fowl. Poultry Sci. 24:155.

Simms, B. T.: 1946. (Poultry Improvement Plans Aid Efficiency of Production.) Report of the Chief of the Bur. of An. Ind., p. 26.

Stafseth, H. J., and Corbutt, A. C.: 1940. Identification of *Salmonella pullorum* colonies with immune serum by means of a macroscopic plate test. Am. Jour. of Vet. Res. 1:76.

Tittsler, R. P., Heywang, B. W., and Charles, T. B.: 1928. The occurrence and significance of *Salmonella pullorum* in eggs. Pa. Agr. Exper. Sta., Bul. 235.

van Heelsbergen, T.: 1929. Handbuch der Geflügelkrankheiten und der Geflügelzucht. Ferdinand Enke, Stuttgart. 104–34.

Van Roekel, H.: 1931. Eleventh annual report on eradication of pullorum disease in Massachusetts. Mass. Agr. Exper. Sta., Control Series Bul. 58.

———: 1935. A study of variation of *Salmonella pullorum*. Mass. Agr. Exper. Sta., Bul. 319.

——— and Bullis, K. L.: 1937. Salmonella infections in chickens. Jour. Am. Vet. Med. Assn. 91:48.

———, Bullis, K. L., Flint, O. S., and Clarke, M. K.: 1932. Twelfth annual report on eradication of pullorum disease in Massachusetts. Mass. Agr. Exper. Sta., Control Series Bul. 63.

———, Bullis, K. L., Flint, O. S., and Clarke, M. K.: 1935. Fifteenth annual report on eradication of pullorum disease in Massachusetts. Mass. Agr. Exper. Sta., Control Series Bul. 78.

———, Bullis, K. L., Flint, O. S., and Clarke, M. K.: 1937. Maltose-fermenting S. *pullorum* strains. Mass. Agr. Exper. Sta., Ann. Rep., Bul. 339:87–90.

———, Bullis, K. L., Flint, O. S., and Clarke, M. K.: 1941. Mass. Agr. Exper. Sta., Ann. Rep., Bul. 378:103.

Villani, S.: 1937. Sulla recettività di alcune specie di volatili all'infezione sperimentale da *B. pullorum*. Profilassi, 10 (4) :148.

Ward, A. R., and Gallagher, B. A.: 1917. An intradermal test for *Bacterium pullorum* infection in fowls. U.S.D.A., Bul. 517.

Weldin, J. C., and Weaver, H. J.: 1930. Transmission of pullorum disease from chick to chick. Poultry Sci. 9:176.

W. R. H.: 1928. New England Conference of Laboratory Workers in Bacillary White Diarrhea Control. Jour. Am. Vet. Med. Assn. 73:263.

## PARATYPHOID INFECTIONS

By R. FENSTERMACHER, *Diagnosis Laboratory,*
*University Farm, St. Paul, Minnesota*

☆ ☆ ☆

Paratyphoid infections in fowl have become problems of considerable importance to the poultry industry in recent years. The production of turkeys has assumed major proportions in the poultry industry in the past few decades; approximately three million turkeys were raised annually from 1910 to 1920. This number has been increased tremendously during the past few years to the extent of more than thirty million birds. It is obvious from the rapid development of the turkey industry that the turkey breeders are especially interested in these problems, as the success or failure of their enterprise often depends upon the control and eradication of these infections. Paratyphoid infections also present problems to the operators of pet bird stores, managers of game bird preserves, and superintendents of parks and zoological gardens where rare birds, animals, and reptiles are maintained. Other interests too numerous to mention at this time are also vitally concerned with these problems.

At the present time, there are more than 150 types of the Salmonella that are paratyphoids. There are two types of Salmonella that do not belong to the group known as paratyphoids; they are *Salmonella pullorum,* the causative organism of pullorum disease, and *Salmonella gallinarum,* the etiological agent of fowl typhoid. All others of the Salmonella types are commonly spoken of as paratyphoids. The different Salmonella types can be distinguished from each other by laboratory tests. The typing and definite identification of the many types of Salmonella, that have been isolated by bacteriological means, have been satisfactorily worked out by Edwards and Bruner of the University of Kentucky. Because of the difficulties involved in typing the organisms of this group, a central laboratory to check the identification is essential for uniform results.

Salmonella organisms cause intestinal disorders in many animals including man. The so-called food poisonings in man in many instances are due to infection by members of this group. Young mammals and birds are much more susceptible to this infection than are mature individuals. It is estimated that approximately 50 types of the total 150 Salmonella types have been

recognized as the cause of enteritis in birds. Forty types of Salmonella have been isolated from turkey poults at the Diagnosis Laboratory, St. Paul, Minnesota, and were found to be responsible for losses to flock owners. Mortality in outbreaks varies greatly. Some of the types do not cause as much loss as others, especially if the poults are under good management. The carrier problem of the recovered or surviving birds is of extreme economic importance, particularly if they are selected as breeding birds. The value of a central typing laboratory to determine the type of infection involved cannot be overemphasized when effective control methods are essential.

Bruner and Edwards (1941) examined approximately 900 cultures which were subjected to serological examination and found 64 of this number were members of the paratyphoid group. Of this group, 92 per cent were isolated from fatal infections of fowl. The distribution of types and the birds from which they were recovered are as follows: *S. senftenberg*, 4 times from turkeys and 3 times from chickens; *S. london*, once from chickens; *S. give*, 5 times from turkeys and 4 times from chickens; *S. anatum*, 15 times from turkeys, 6 times from chickens, and 3 times from ducks; *S. meleagridis*, 4 times from turkeys and 1 time from chickens; *S. newington*, 6 times from turkeys, 2 times from chickens, and 1 time from ducks; and *S. new brunswick*, 3 times from turkeys and 1 time from chickens. The remaining 5 cultures were recovered from hogs, man, and water. The type *S. senftenberg* has been recovered from man and is quite frequently recovered from poults. Edwards and Bruner (1943) examined several thousand cultures of Salmonella obtained from many areas in the United States. They did not subdivide the fowl into their respective groups such as turkeys, chickens, and ducks. Forty Salmonella types were found in the group. The paratyphoid types identified and the number of outbreaks are listed in Table 1.

Multiple type infections are common and have been reported by many

TABLE 1

| Salmonella Types | No. of Outbreaks | Salmonella Types | No. of Outbreaks | Salmonella Types | No. of Outbreaks |
|---|---|---|---|---|---|
| aberdeen | 4 | hivittingfoss | 4 | oregon | 2 |
| amherstiana | 1 | illinois | 2 | panama | 6 |
| anatum | 44 | kentucky | 6 | paratyphi B | 4 |
| bareilly | 47 | litchfield | 1 | rubislaw | 1 |
| bredeney | 27 | london | 1 | saint paul | 1 |
| california | 16 | manhattan | 2 | san diego | 3 |
| cerro | 2 | meleagridis | 15 | senftenberg | 15 |
| cholera-suis | | minnesota | 4 | thompson | 1 |
| var. kunzendorf | 2 | montevideo | 11 | typhimurium | 409 |
| derby | 33 | muenchen | 2 | typhimurium | |
| dublin | 1 | new brunswick | 26 | var. copenhagen | 63 |
| eastbourne | 2 | newington | 18 | urbana | 1 |
| enteritidis | 5 | newport | 19 | wichita | 1 |
| give | 18 | oranienburg | 28 | worthington | 18 |

research workers. Pomeroy and Fenstermacher (1941) reported multiple type infections in turkeys. Other reports may be found in the literature indicating similar occurrences. From one farm which obtained poults from several sources, the following types were isolated: *S. pullorum, S. typhimurium, S. derby, S. give, S. oranienburg*, and *S. anatum*. In previous years from the same farm, *S. worthington, S. bareilly*, and *S. enteritidis* had been recovered. From another farm which received poults from one hatchery, cultures of *S. pullorum, S. chester, S. derby, S. anatum*, and *S. oranienburg* were isolated.

Specific reference is made later to the work of Hinshaw and his colleagues on the occurrence of paratyphoid organisms recovered from reptiles and other zoo animals. The presence of paratyphoids in reptiles is another important factor of concern to turkey breeders located in areas where snakes are of common occurrence. Paratyphoid organisms have also been isolated from rats and mice.

Man must also be recognized as a possible source of infection. This may involve not only the caretakers but also the waste products of human source. Hinshaw, McNeil, and Taylor (1944) record instances of human sources of infection to poults and also the transmission of two types of paratyphoids (*S. panama* and *S. montevideo*) to man believed to have occurred as a result of handling infected poults.

Darby and Stafseth (1942) reviewed the literature with reference to the thirty-five species of the genus Salmonella found in poultry in the United States. The literature incriminating most of these species in pathological conditions in man has been cited. The relation of Salmonella occurring in poultry to public health has not as yet been determined; however, flies can definitely be incriminated as vectors of paratyphoid organisms.

**Symptoms** alone are not specific enough to make a definite diagnosis, with the possible exception of the swellings that are observed near the wing joints of pigeons. Usually the only clinical symptoms observed are that sick birds stand around with wings drooping and feathers or down ruffled, showing evidence of diarrhea, pasting of the vent, and a tendency to huddle near the heating element. Specific symptoms have been included under each species of bird mentioned in the chapter.

**Lesions** are not always found. A number of the birds should be examined. Diagnostic laboratories prefer to have about six birds for examination, part of which should be live birds showing well developed clinical symptoms and the others should be dead specimens which have been properly refrigerated for shipment or delivery. The young birds, if affected with an acute type of the disease, generally show very few lesions or none at all. If lesions are found, they generally are not sufficient to make a positive diagnosis. If the disease is more chronic in type, the most common lesions will be an enlarge-

ment of the liver and sometimes the spleen, and small necrotic foci may be seen on the surface of the liver. Cecal cores are suggestive of paratyphoid infection but they may be the result of pullorum disease. Sometimes the air sacs are cloudy. The latter is also found in birds of all ages (chickens and turkeys) affected with Newcastle disease. Further pathologic changes will be described under the different species included in the chapter.

**Differential diagnosis** is difficult and cannot be made without the aid of a competent diagnostic laboratory. The diseases that must be kept in mind are pullorum disease, respiratory infections, Newcastle disease, and accidents that occur during brooding.

**Diagnosis** depends upon the isolation and identification of the organism recovered from the organs and intestinal contents. This means, of course, that the practitioners of veterinary medicine should request the assistance of a reputable laboratory. The diagnostician no longer can depend upon the results of the bacteriological examination of the heart blood and liver especially when the poults are being examined. Frequently those organs fail to yield paratyphoid organisms, and recoveries are made by cultural examination of the intestinal contents. It is not necessary in order to establish a diagnosis of paratyphoid infection, for the laboratory making the examination to do the serotyping of the organism or organisms isolated, until a control program for paratyphoid infections is undertaken.

**Bacteriological examination** technique varies, depending upon the laboratory doing the work. It makes little or no difference as to the variations used as long as recognized methods are adopted. When making daily examinations of specimens submitted for the purpose, a suitable method consists of opening the body cavity of the bird as aseptically as possible and exposing the internal organs. The usual precautions are taken by searing the surface of the organs to be examined with a hot spatula. The cultures are seeded either on plain broth agar enriched by adding 5 per cent horse serum or on dextrose starch agar. Small portions of intestinal contents are added to tubes containing 8 to 10 cc. of tetrathionate broth to which has been added brilliant green (1:50,000). Selenite F (Difco) is also used for culturing intestinal contents. The cultures are incubated for 24 hours at 37.5° C. At the end of 24 hours of incubation, colonies resembling Salmonella are transferred to another slant (either serum agar or dextrose starch agar) and again incubated 24 hours. If growth is abundant and not contaminated as result of the original seeding, the typical colonies are transferred to the following carbohydrates: dextrose, lactose, sucrose, mannite, and maltose. These are incubated and observed at the end of 24 hours. If the selected organisms develop acid and gas in dextrose, mannite, and maltose, they are considered a paratyphoid. Serotyping will definitely identify the type or types involved. S S plates are seeded with a loopful of the intestinal cultures. At the end of 24

hours of incubation, the S S plates are examined, and typical Salmonella colonies are transferred to agar slants and incubated another 24 hours. The procedure from this point is as above for the cultures obtained from the internal organs. If the laboratory worker has sufficient assistance, a dozen or more typical colonies should be picked from the plates. In this way more Salmonella types would probably be isolated. The intestinal cultures are usually taken from the duodenum, the mid-gut, and the rectum.

Adult birds, especially turkeys, that are submitted for bacteriological examination as the result of being reactors to serological tests are submitted to a more careful and exacting examination as outlined by Pomeroy and Fenstermacher (1939).

**Control** of paratyphoid infections of turkeys is of great economic importance. There is little doubt that the disease is becoming more widespread among chickens. It is also of importance to the pigeon fancier. The most practical way to control the disease among birds in the pet or bird stores is by means of depopulation and thorough cleaning and disinfection of the premises. A critical examination should be made of the source of supply for replacements in order to prevent recurrent infection.

A gradual decline of pullorum disease among turkeys has been recorded during three years of observation. During this same period, a marked increase in the paratyphoid infections has occurred (Table 2).

Every possible effort to control and eradicate paratyphoid infection from a breeding flock of turkeys should be made. Control of the disease should not be the only objective. As long as a single carrier remains in the flock, there is potential danger of another outbreak among the poults. Therefore,

TABLE 2

INCIDENCE OF SALMONELLA INFECTIONS AND MORTALITY RECORDS OBTAINED FROM NATURALLY INFECTED FLOCKS OF CHICKS AND POULTS WITH VARIOUS TYPES OF SALMONELLA

| Host and Disease | Year | Total No. of Flocks Examined | Percentage Flocks Infected | No. of Infected Flocks Reported | No. of Birds Involved | No. of Birds Died | Percentage Loss in Infected Flocks |
|---|---|---|---|---|---|---|---|
| *Chicks* | 1944... | .......... | .......... | 126 | 63,207 | 21,656 | 34.1 |
| Pullorum | 1945... | .......... | .......... | 144 | 61,658 | 13,600 | 22.1 |
| disease | 1946... | .......... | .......... | 167 | 67,582 | 23,093 | 34.1 |
| *Poults* | 1944... | 275 | 23.2 | 49 | 125,336 | 26,772 | 21.3 |
| Pullorum | 1945... | 288 | 20.2 | 30 | 48,039 | 9,256 | 19.3 |
| disease | 1946... | 283 | 7.8 | 14 | 34,413 | 6,285 | 18.2 |
| Paratyphoid | 1944... | 275 | 25.4 | 42 | 74,695 | 18,580 | 24.3 |
| infection | 1945.. | 288 | 30.0 | 40 | 115,325 | 11,479 | 10.0 |
|  | 1946... | 283 | 41.5 | 76 | 120,561 | 26,466 | 22.4 |
| Mixed: | 1944... | 275 | 16.4 | 28 | 69,991 | 21,567 | 30.4 |
| pullorum and | 1945... | 288 | 11.1 | 17 | 24,970 | 5,076 | 20.4 |
| paratyphoid | 1946... | 283 | 5.8 | 9 | 21,064 | 5,272 | 25.0 |

poults that die during the brooding and rearing season should be examined bacteriologically; and if paratyphoids are isolated, these must be typed to determine the particular Salmonella causing the infection. Multiple type infections are indeed common. There have been as many as seven different paratyphoids found among the poults on a single farm or ranch. If multiple types are found, there is little hope of success in eradicating the disease among the breeding stock by agglutination testing. The O and H antigens must be prepared from each Salmonella type present in the flock and each bird subjected to repeated agglutination test with whatever types are known to exist in the flock. The most economical method is to obtain foundation stock from sources not affected with any of the paratyphoids. There is a possibility of controlling paratyphoid infection among turkeys where no more than one type is present, but experience has shown that this may be more difficult than anticipated. Since vermin, reptiles, and flies are known to be vectors of Salmonella, these will introduce additional factors that must be considered in the control of the disease. Flies are an ever present menace.

**Treatment** is not satisfactory. Many drugs and combinations of drugs have been tried, and all have failed to prove satisfactory. With the advent of the sulfonamides, there was hope that some one or a combination of them would provide an effective agent for control of these infections until more practical control methods were developed. Pomeroy and Fenstermacher (1946, unpublished data) fed sulfadiazine, sulfamerazine, and sulfamethazine to chicks experimentally infected with *S. typhimurium*. Losses were reduced, but results indicated the need for further study. Sulfathiazole, sulfaguanadine, and sulfadiazine were fed to poults artificially infected with *S. typhimurium*, and the results were less effective than when fed to chicks. The reason for this difference is not definitely known but is thought to be associated with the difficulty of getting the poults to eat when the drugs were added.

**Microscopic pathology.** The lesions found in birds affected with paratyphoid infection are not characteristic enough for positive diagnosis. Many tissues have been examined that were obtained from poults known to have died as a result of this infection, and it is probable that a diagnosis, if made in that manner, is likely to be incorrect in many instances.

## PARATYPHOID IN CANARIES AND PARROTS

Beaudette and Edwards (1926a), in 1924, investigated paratyphoid in canaries and parrots. In one bird store, 200 birds of all ages became affected. The mortality was 35 per cent. Another breeder lost 25 birds out of a group of 33. The etiological agent was determined to be a member of the paratyphoid-enteriditis group, and at that time the organism was considered

to be identical with the type designated by some authorities as *B. aertrycke* and by others as *B. pestiscaviae*.

Emmel and Stafseth (1929) reported a few outbreaks of an epizootic that occurred in canary bird stores throughout the state of Michigan. The disease was highly infectious, and the mortality was high. The incubation period was 4 or 5 days; the course of the disease varied from 2 to 4 days. *S. aertrycke* was isolated from the internal organs.

Fenstermacher *et al.* (1927–46, unpublished data) have on several occasions found that *S. aertrycke* caused the death of many canaries assembled in bird stores. The mortality was always high. If the bird cages were thoroughly cleaned and disinfected following an outbreak, the cages could be safely restocked with healthy birds. However, new birds coming in contact with survivors usually became infected.

Beaudette (1926a) reported that *S. aertrycke* caused the death of parrots as well as canaries. Aside from the fact that about fifty birds were known to have died, the history was scanty. Beaudette could not differentiate between the strains of *S. aertrycke* isolated from the parrots and those previously isolated from canaries.

Altman (1940) studied an outbreak of a disease among a group of 170 canaries of all ages. The incidence of the infection was greater in the young birds. Likewise, the heaviest mortality was among the young birds. Approximately 60 per cent of the infected birds died. *S. suipestifer* was isolated as the etiological agent.

**Symptoms:** The affected birds seemed to "puff up," developed convulsions, and died within 2 or 3 days. Constipation was observed in some cases; later, diarrhea developed. The droppings were greenish in color and, in some instances, bloody. Large amounts of urates appeared in the feces shortly before death.

**Post-mortem** lesions consisted of catarrhal enteritis, with a congestion of the mucosa; intestinal contents frequently were bloody; liver and lungs showed congestion; the kidneys were congested, with distension of the ureters; the spleen was enlarged to three or four times normal size; and the myocardium was dark red in color.

**Treatment** was attempted by means of an autogenous bacterin injected in 0.1 cc. and 0.2 cc. doses. The 0.2 cc. doses appeared to be the maximum amount tolerated with safety. The vaccinated birds and the controls all remained well when vaccinated several weeks following the subsidence of the outbreak.

### PARATYPHOID IN CHICKENS AND GUINEA FOWL

There is little doubt that paratyphoid infections among chickens have existed for many years. Unfortunately, the early research workers did not

have present day methods available by which the isolated organisms could be positively identified. Mazza, in 1899, was one of the first to describe a chicken epizootic that raged in various parts of northern Italy. He isolated an organism that was motile, gave a negative indol test, coagulated milk, fermented glucose, and was pathogenic for chickens and pigeons, but not for rabbits. This organism might have been a paratyphoid. The records of the Diagnosis Laboratory at St. Paul, Minnesota, indicate that it was not until 1929 that *S. typhimurium* was isolated from the internal organs of twelve chicks. Other research workers prior to that time and many more during the past two decades have encountered the disease among chickens. There is no doubt that the infection does not occur in chicks as frequently as in turkey poults. However, the incidence of this infection in chicks has increased and threatens to become a greater economic problem.

Pfeiler and Rehse (1913) recovered from a cock an organism which was morphologically and serologically identical with the paratyphoid B bacillus. The organism was not pathogenic for chickens, pigeons, or ducks. Since only one loss occurred in the flock, it was concluded that this was a question of an individual case of a disease due to paratyphoid B bacillus. Lütje (1921) observed a very interesting case of paratyphoid in chickens caused by a paratyphoid B bacillus of the Smith-Kilborne type. Spray and Doyle (1921) described an organism of the paratyphoid B group which was isolated from baby chicks. Twenty strains of paratyphoid organisms were isolated. No correlation could be shown in gas production in the various carbohydrates. Some strains produced gas in glucose and not in mannite; the behavior of others was exactly the reverse. In order to obtain gas production, it was necessary to repeat the trials. Edwards (1929) has stated that organisms of the paratyphoid B group have often been found in infections of fowl. Caged birds were affected more frequently than the barnyard fowl. The close resemblance of paratyphoid infection of baby chicks to pullorum disease is mentioned. Organisms responsible for the disease were regularly recovered from the liver, heart, lungs, and unabsorbed yolk. In the outbreak studied, Edwards determined by means of agglutination tests that the breeding stock was free of the disease. It is probable therefore that these chicks became infected after hatching, even though the early appearance of the disease suggested transmission by means of the egg. This contention was later supported by other investigators. Two organisms of the paratyphoid group, *S. typhimurium* and *S. anatum,* were isolated from the chicks examined. This constitutes the first report of the presence of the latter organism in chickens; the former was known to be widespread. Doyle (1927) in 1925 investigated a disease among chicks wherein the entire hatch of chicks had died and 50 per cent of another hatch had also succumbed by the fifth day. A heavy mortality (69 per cent) had also occurred among young ducks, though none

of the latter were submitted for examination. *S. typhimurium* was regularly isolated from the internal organs of the chicks. The origin of the infection was not determined, although contaminated material might have been the means of introduction. Gaiger and Davies (1933) later investigated another outbreak of a paratyphoid disease among baby chicks in England. In this case the eggs originated in Scotland and were hatched on a poultry farm located in southern England. Sickness was observed about the tenth day following hatching, and *S. typhimurium* was recovered from the internal organs of the dead chicks. It was believed that the infection was introduced with the eggs. Schalm (1937) investigated the outbreak of a disease among chicks caused by an organism closely resembling *S. aertrycke*. The breeding flock which produced the chicks had been under his supervision for three years prior to the outbreak. During this time, neither paratyphoid nor pullorum organisms had been isolated from either chicks or adults subjected to a bacteriological examination, nor had any pullorum-disease reactors been found when tested by the whole-blood method. Chicks were hatched in forced draft incubators throughout the year. Losses during the first part of April varied from 6 to 40 per cent of the chicks which were sold up to three weeks of age. A pure culture of an organism belonging to the paratyphoid group was isolated from five different groups of chicks submitted for examination. There was a great contrast in the small loss among the chicks kept on the farm where hatched, in comparison to the heavy mortality among the chicks sold to others and transported to new quarters. It was believed that the resistance of the chicks which were sold might have been lowered by holding them overnight in boxes containing 100 chicks each. A rather powerful ventilating fan had been installed in the storage room, and possibly those chicks nearest the fan became chilled during the night. Since rats had become a nuisance in the feed room, a dozen rats were examined bacteriologically, but with negative results. The evidence indicated that the chicks received the infection in the incubator through the respiratory organs after hatching, rather than through the egg. It was necessary to determine the source of the incubator infection. It was possible that some of the breeding stock harbored paratyphoid organisms in their intestinal tract; if this were so, the outside of the eggshell could easily have become contaminated. Pieces of eggshell were sterilized and then smeared lightly with sterilized chicken feces to which a broth culture of the paratyphoid bacillus had been added. These eggshells were then stored in an incubator at 37° C. Living organisms were found 111 days later. Schalm further demonstrated that the paratyphoid bacillus is capable of penetrating the shell and infecting the developed embryo when smeared on the outside of eggshells prior to incubation. It is possible that the infectious agent was introduced into the respiratory tract after hatching. A group of eighteen chicks was fed viable paratyphoid

organisms in doses varying from one to three drops of an 18-hour broth culture. Five of the group died between the fourth and fourteenth day following the administration of the culture. The internal organs and intestinal contents of each were cultured on brilliant green agar with negative results. The survivors were killed on the nineteenth day and examined bacteriologically with negative results.

The earliest paratyphoid infection of chickens found in the records of the Diagnosis Laboratory, St. Paul, Minnesota, occurred in 1929. Later infections occurred as follows: 1930, two chicks; 1933, four chicks; 1934, four chicks; 1935, seven chicks; 1936, four chicks; 1937, three chicks; 1938, four chicks; 1939, four chicks; 1940, thirteen chicks; 1941, three chicks; 1942, eight chicks; 1943, ten chicks; 1944, nine chicks; and 1945, twenty-four chicks. The actual mortality that occurred as a result of the disease is not definitely known.

Jungherr and Clancy (1939) isolated paratyphoid organisms from fifteen cases out of a group of 1,241 specimen consignments of chicks. These specimens were received during the period from 1930 to 1938. Ten of these cases originated from stock listed as "pullorum clean." Where follow-up examinations were made, the same serotype was found as in the first isolation. This is a significant finding in view of the multiple serotypes to which the common fowl is susceptible. The affected chicks varied in age from 4 to 21 days. In fourteen cases which were included in the serologic study, four of the isolated organisms were classified, according to the Kaufmann-White scheme, as *S. typhimurium*, two as *S. typhimurium* var. *binns*, one as *S. bareilly*, two as *S. oranienburg*, three as *S. montevideo*, one as *S. london*, and one as *S. anatum*.

Van Roekel and Bullis (1937) have discussed the difficulties that are encountered in the testing of chickens for the control of pullorum disease when some of the paratyphoid infections are present in flocks. Certain flocks, when retested repeatedly, are found to have a few birds that react each time when tested by pullorum test tube antigen. Cross agglutination frequently occurs with other members of the Salmonella group, limiting the specificity of the pullorum test. The complication of the control of pullorum disease in this instance was due to the presence of a paratyphoid organism (the type had not been determined). There were at least 2 per cent additional reactors disclosed. Paratyphoid organisms were recovered from a number of the birds that were examined bacteriologically. Edwards (1938) isolated *S. kentucky* from the intestinal tract of a chick affected with coccidiosis and ulcerative enteritis. The organism possessed two previously undescribed antigens, one somatic, the other flagellar. Edwards and Bruner (1938) identified a paratyphoid organism that Pomeroy isolated from a chick as a new type and is referred to as *S. minnesota*. Gordon and Buxton (1945) have isolated *S.*

*thompson* on forty-four occasions from thirty-one outbreaks of paratyphoid infection among chicks and from two outbreaks among ducklings. The mortality among chicks varied from 20 to 80 per cent, and in one hatch a 100 per cent loss occurred. Hoffman *et al.* (1943) stated that 97.7 per cent of the cases of paratyphoid were in chicks less than five weeks old.

Hudson (1942) encountered in 1941 an outbreak of paratyphoid infection among a flock of guineas whose owner reported a loss of sixty birds during the preceding six weeks. *S. bredeney* was isolated from the infra-orbital sinuses.

**Symptoms** observed consisted of drooping wings, huddling, discharge from the eyes and nostrils, and slobbering from the mouth. Occasionally the head was thrown back, and sometimes the affected birds would close or clench their claws.

**Post-mortem.** A more or less clear mucous exudate could be expressed from the nasal cleft and external nares by applying external pressure in the region of the sinuses. The sinuses were filled with a yellowish, mucoid exudate.

**Bacteriological examination.** A pure culture of a paratyphoid organism was isolated from the infected birds. This organism was later identified as *S. bredeney* by Edwards at the University of Kentucky.

Hinshaw, Taylor, and McNeil (1942) isolated *S. bredeney* and *S. typhimurium* from a group of chukar chicks in 1939. This report clearly indicates the isolation of multiple Salmonella types from a single outbreak wherein a 48.2 per cent mortality occurred among a group of 1,061 chukar chicks.

**Symptoms** shown by the affected birds are not typical enough to make a definite diagnosis of paratyphoid infection. The adult birds may be carriers of infection. Some of the affected chicks may be found dead without showing any definite clinical symptoms. The principal symptoms manifested include inclination to huddle near the heating element, drowsiness, ruffled feathers, diarrhea, pasty vents, and frequently, difficult respiration. As a rule the disease makes its appearance at two different age periods. The first is when the chicks are 4 to 5 days old and the other when they are 10 to 12 days old. The peak of losses of the first age group is generally reached by the tenth or twelfth day. The peak of mortality among the second age group occurs when they are about two and a half to three weeks old. The mortalities that occur in the two age groups are probably due to the amount of infection present in the incubator at the time of hatching. If there is a heavy infection present at that time, it is likely that the heaviest losses will occur early. Lack of strict sanitary methods in rearing the chicks increases the incidence of the disease. The losses vary from 10 to 80 per cent or even higher.

**Post-mortem** lesions are not distinctive. The lesions caused by paratyphoid types in chickens cannot be differentiated from those caused by

*S. pullorum.* Diarrhea and retained egg yolk, as a rule, are not common findings. Epicarditis and pericarditis are more frequently observed. Occasionally the pericardial fluid is yellow, containing a fibrinous exudate which may be adherent to both the epicardium and pericardium. Exudates are infrequently found on the surface of the liver and the lungs. Occasionally the ceca may be filled with firm, pale yellowish plugs. Frequently the autopsy findings are likely to be negative especially if only one or two birds are examined. The internal organs of the adult carrier bird generally present no lesions that are characteristic of paratyphoid infection.

**Diagnosis** is made either by means of bacteriological examination and the isolation and identification of the organism or by means of the agglutination test, using both O and H antigens of the Salmonella type known to be present in the flock. The latter should be determined by means of examination of chicks that die during the brooding stage.

### PARATYPHOID IN FINCHES AND SPARROWS

Manninger (1913), in 1912, examined the intestinal flora of various birds that had died suddenly. Among the birds were siskin, gold finches, and green finches. An acute intestinal catarrh was observed. In three birds belonging to the family of finches, he recovered a paratyphoid B bacillus from the intestinal flora. The same organism was recovered from the internal organs and blood of nine other birds.

### PARATYPHOID OF GEESE AND DUCKS

Young geese and ducks are susceptible to paratyphoid infection. Outbreaks often become epizootic. The disease is often spoken of as "keel" disease in ducklings. Manninger (1918) described the disease as avian paratyphoid and succeeded in isolating a paratyphoid B type organism which he considered the etiological agent. Pfeiler (1920) observed one outbreak of paratyphoid in older geese. The flock contained fourteen geese, nine weeks old; ten of the geese died, and the others were ill. A motile bacillus was isolated that closely resembled the mouse typhoid organism, not only culturally but also serologically. Weisgerber and Müller (1922) observed extensive losses among young geese in East Prussia. Again a paratyphoid organism was considered as the etiological agent. Rettger and Scoville (1920), in 1918, studied a disease that affected young ducklings. The loss was almost 100 per cent among a lot of 3,000 ducklings. The heaviest losses occurred during the first 10 days after hatching. *S. anatum* was isolated from the internal organs of the young ducklings. The culture was later examined by Edwards and Rettger (1927) and found to contain two distinct types, *S. aertrycke* and *S. anatum* (Edwards, 1935a, b). Clarenburg (1939) considers paratyphoid in ducks of foremost importance in connection with public health. Eggs may

be infected as a result of localization of the organism in the ovary, and since the organisms are excreted with the feces, the outside of the shell may also become contaminated. Hole (1932) investigated a number of epizootics among ducklings. While studying three outbreaks, he recovered an organism apparently identical with *S. enteritidis* in one case, and an organism closely allied to *S. aertrycke* in the other two cases. Evidence was presented indicating the possibility of egg transmission.

Little is known regarding the occurrence of paratyphoid infection in wild ducks. Levine and Graham (1942) examined ten dead wood ducks about one week old. The eggs were obtained from wild ducks by using nesting boxes. The eggs were hatched in an incubator that had never been used. Ninety-four per cent of the eggs hatched, and the ducklings were reared in cleaned and disinfected pens. Approximately 400 out of 500 ducklings died. *S. typhimurium* was isolated from the heart blood and livers of a number of the ducklings examined bacteriologically. Fecal samples were collected from sixteen surviving, healthy appearing wood ducks. *S. typhimurium* was recovered from two ducks and *S. bredeney* was recovered from one other duck as a result of examining the feces bacteriologically. Other wild birds were present on the preserve, and feces were collected from five mallards and three Canada geese. Blood samples were taken from nine mallards and fourteen Canada geese. The feces were cultured and *S. typhimurium* was isolated from one mallard. The sera were tested by test-tube and rapid agglutination tests in a dilution of 1:25. All were negative to the tube test, but with the rapid serum test a positive reaction was obtained in the mallard from which Salmonella was isolated. Garside and Gordon (1943–44) investigated an extensive outbreak of salmonellosis in ducklings. *S. typhimurium* and *S. enteritidis gaertner* were recovered from the liver and pericardial fluid from a fairly representative number of the birds examined. The losses among the 57,000 ducklings on the premises during the first month of their life was approximately 30 per cent. Two peak periods of mortality were experienced; the first occurred 3 to 4 days after hatching and the second at 12 to 14 days of age. Losses ceased after removal from the brooder house on the twenty-eighth day.

**Symptoms** manifested by geese are not pronounced, and the literature contains little regarding clinical symptoms. Loss of appetite is usually observed, followed by weakness and loss of condition. Frequently the eyelids become swollen or edematous. Some of the infected individuals develop a diarrhea. Infected ducklings which die within a few days after hatching seldom manifest clinical symptoms. Older ducklings may become weak and sluggish. They hover about the heating element, are not easily aroused, and lack energy to search for food. Intense thirst is usually developed. The affected birds stand upright after drinking, "keel over," and after several gasps, die; hence, the name "keel" disease was designated by Rettger and

Scoville (1920). Sometimes the head is drawn back until the beak rests dorsally on the spine. Other times they may fall over backward or to the side, kick violently, occasionally twisting the head and neck in circles.

**Post-mortem** lesions are usually not characteristic and are of little aid in making a specific diagnosis in ducklings 3 to 4 days old. There may be congestion of the lungs or retention of the yolk sac, but these are not specific for paratyphoid infection. Ducklings two to three weeks old may show more definite lesions. The most consistent pathologic change is the mottled appearance of the liver, irregular areas of congestion, and enlargement of the organ. The kidneys may also show congestion and enlargement. Sero-fibrinous pericarditis is often observed.

**Diagnosis** must be confirmed by bacteriological means. The examination of the feces should not be overlooked as the infectious agent is frequently isolated from this material.

**Control** of this disease by the present accepted methods is not entirely satisfactory. Attempts can be made by testing the blood sera of the breeding stock by test-tube agglutination method, using O and H antigens. This method will not satisfactorily detect the carrier birds, but conditions may exist which justify the use of this method. Fecal examinations can also be made. The establishment and rigid execution of an effective sanitary program is essential in the control of all infectious conditions.

### PARATYPHOID IN PIGEONS

The first authentic report of pigeon paratyphoid was made by Mohler (1904). Moore (1895) investigated a disease that affected squabs and, occasionally, old pigeons. He isolated an organism that was thought to be a variant of the hog cholera bacillus. Beaudette (1926b), in reporting this disease in squabs, mentions that the outbreak which he investigated had occurred in the southern part of New Jersey as had those reported previously by Moore and Mohler. Gauger *et al.* (1940) published a comprehensive study of pigeon paratyphoid. The etiological agent was *S. typhimurium* var. *binns.* Niemeyer (1939) recovered an organism from pigeons that was identified by Edwards as *S. typhimurium.* In this outbreak, *Trichomonas columbae* was concurrently present. The losses of both adults and squabs in the loft were heavy. The paratyphoid organism was recovered from three of the fourteen pigeons examined.

Jungherr and Wilcox (1934) reported on a variant *S. aertrycke* recovered from spontaneously infected squabs that were negative to tube agglutination tests. Others have had similar results. The organism is frequently recovered from birds negative to tube agglutination tests in which both live and phenolized antigens are used. Low-titer birds are not detected by the rapid whole-blood agglutination tests.

Edwards (1935b) subjected the variants of *S. aertrycke* isolated by Jungherr and Wilcox, as well as by Black and by Edwards, to a more critical study. The cultures differed from *S. aertrycke* in that they possessed somatic antigens identical to those of *S. abortus equi*. The type has been designated as *S. aertrycke* var. *storrs*.

Hoffman and Edwards (1937) isolated paratyphoid bacilli from the internal organs of three pregnant rabbits as well as from their fetuses. Paratyphoid infection of rabbits is rather uncommon. Three pigeons of a group that were housed close to rabbits were examined. A Salmonella was isolated from one with an enlarged humeroscapular joint and was identified as IV variant of *S. aertrycke*. It is quite probable that it was transmitted from the pigeons to the rabbits. The European literature contains many references to *S. aertrycke* infections in man which were traceable to ingestion of eggs of infected pigeons. According to Edwards, the IV variant of *S. aertrycke* isolated from pigeons is quite universal in distribution.

A report on pigeon paratyphoid was published by Lahaye and Willems (1927). They considered paratyphoid infection to be one of the most important diseases of pigeons in Belgium. During 1930 Khalifa (1935) investigated an epizootic among pigeons in Cairo, Egypt, which was due to *S. typhimurium*. The cultural and biochemical reactions of the organism isolated were recorded. Guinea pigs were found susceptible. Morcos (1935) in 1932 also studied an outbreak among pigeons in various lofts in Cairo. The organism isolated was found closely allied to *S. anatum*. Adult fowls and sparrows were found to be quite refractive to the isolated culture. Shirlaw and Iyer (1937) encountered an unusual loss among pigeons that were being used for the production of fowl pox vaccine. The organism isolated was *S. enteritidis*, which was subjected to extensive agglutination, cross agglutination, and agglutination absorption tests.

'Symptoms. Adult pigeons may or may not show evidence of being infected. The majority of them, however, show no symptoms. Emaciation, if present, is generally well marked in chronic cases. Frequently the evacuations are loose and have a green color. Occasionally the eyes are glassy and watery, and the mucosa is congested. Arthritis is not commonly observed but when present generally involves the wing joints, and occasionally other joints are affected. Soft swellings, subcutaneous in location and near the affected joints, are often present. These lesions may appear in squabs two to three weeks old. Similar swellings may be found on other parts of the body. Abnormal losses among squabs may be the first indication of the presence of the disease. Squabs are often emaciated, unable to stand, droppings similar to adults, and the eye symptoms are generally more prominent.

Post-mortem. The soft swellings contain a fluid which is generally of a serous type. Occasionally this fluid appears yellowish and becomes thicker

in consistency. The internal organs may not show any marked lesions. The principal lesions consist of enlargement and congestion of the liver, spleen, and kidneys. The ureters are distended with urates. Small necrotic foci of the liver develop in the chronically affected birds. Small necrotic foci are more frequently found in the lungs than in the liver. Air sac walls may show thickened areas. These may appear like flecks or granular areas that are cheeselike in consistency. The ova may appear abnormal, frequently the surfaces are markedly congested. Encapsulated masses, varying in size, containing egg yolk or yolklike material are often found attached to various internal organs. Enteritis is a more constant lesion especially in those cases in which intestinal infection occurs. Slightly raised, grayish patches of the intestinal mucosa are observed quite frequently.

**Diagnosis**. In order to make an accurate diagnosis, it is necessary to subject the internal organs and intestinal contents to careful bacteriological examination. Methods of procedure have been outlined in the earlier portion of this chapter. Attention is called to the fermentation of maltose which is often quite slow. Acid and gas are usually formed in this carbohydrate, but occasionally no acid and, more frequently, no gas is formed. This irregularity in cultural characteristics is mentioned so as not to confuse the paratyphoid organism with the nonmotile *S. pullorum* organism.

**Transmission**. It is definitely known that the causative organisms may be transmitted to young pigeons by infected adults in the same manner as pullorum disease. The organism may be recovered from eggs. It is also very commonly found in the pharyngeal fluid of infected adults, and infected pigeons may void viable organisms in their feces. Paratyphoid organisms are present quite commonly in the blood, mouth fluids, and feces of young squabs showing symptoms of paratyphoid infection.

**Control** of the disease is difficult. Rapid whole-blood antigens prepared from cultures of *S. typhimurium* are not satisfactory diagnostic agents. Repeated tube agglutination tests are usually too expensive to be of practical value. The O and H antigens should be prepared from the autogenous culture causing the outbreak. Gauger *et al.* (1940) found that eight months after an epizootic among 636 pigeons, not including the nestlings, 37 per cent agglutinated the antigen in dilutions of 1:25 or higher. Studies were made to determine whether pens of birds kept under practical sanitary conditions could be freed of reactors by repeated agglutination tests. After each test the reactors and negative mates to the reactors and nonmated reactors were removed from the building. The results were not encouraging, but if a program of this kind were carried out for a long period of time the disease could eventually be eliminated. Hatchability and livability may be quite satisfactory in some lofts even though the breeding stock may be quite highly infected. The peculiar method by which adult pigeons feed their young

further complicates the problem of control. Thus infected adults can readily transmit the infection to the young.

## PARATYPHOID IN QUAIL

Graham (1936) studied an outbreak of a disease among quail ranging in age from 3 days to six weeks which were being hatched by the State Department of Conservation. Many of them died without showing premonitory symptoms. Baby quail that appeared healthy in the evening were found dead the following morning. In some brooders the mortality was as high as 75 per cent. It was estimated that 60 per cent of all the quail hatched died. Graham cultured fresh eggs as well as the yolk sacs of dead embryos. From these, unidentified streptococci and colon type organisms were recovered. From the livers of five dead quail, *Escherichia coli* and Salmonella-like organisms were recovered. The Salmonella culture was submitted for further identification to Edwards (1936), who found the organism to be *S. oranienburg*.

Cunningham (1941) encountered an acute paratyphoid infection among quail chicks in which the heaviest mortality occurred in chicks from 3 to 9 days old. From 18,580 eggs incubated, 62.5 per cent hatched. Fertile egg hatchability for the season was 82 per cent. A total of 11,516 chicks were placed in the brooder house. Of this number 9 per cent were abnormal. The mortality during the four weeks the birds were in the brooder house was 38.3 per cent. At four weeks of age the birds were sent to the game preserve where losses were negligible. *S. bredeney* was isolated as the etiological agent. Agglutination tests with whole-blood antigens prepared from the isolated strain of *S. bredeney, E. communior, E. communis,* and a standard *S. pullorum* did not reveal the presence of any agglutinins when forty-five mature quail from the laying flock were tested.

**Diagnosis** is dependent upon bacteriological examination.

**Control** of the disease is not satisfactory. Until more accurate means of detecting the carrier birds are developed a more rigid sanitary program should be instituted for the control of the disease.

## PARATYPHOID INFECTIONS IN SNAKES, TURTLES, GILA MONSTERS, IGUANA, CATS AND FLIES

For many years, diagnosticians and research workers were confronted with outbreaks of salmonellosis, or what is more specifically spoken of at the present time as paratyphoid infections, among poults wherein it was impossible to trace the source of the infection.

Hinshaw and McNeil (1945) examined tissues and stool specimens from 153 animals, consisting of 21 mammals, 85 birds, and 47 reptiles on exhibit at the San Diego, California, Zoological Gardens. A total of seven Salmonella

types were isolated including *S. typhimurium, S. kentucky,* and *S. panama.* McNeil and Hinshaw (1946) also isolated *S. san diego* and *S. newport* from Galapagos turtles, *S. montevideo* from a Gila monster, and *S. manhattan* from an iguana. In another study, Hinshaw and McNeil (1945) examined forty-one snakes caught on ranches in seven localities. Eleven of the snakes yielded Salmonella. Four types were isolated: *S. meleagridis, S. typhimurium, S. newport,* and *S. rubislaw.* Garter and gopher snakes were found to be reservoirs of infection. A fifth type, *S. panama,* was isolated from a Cuban snake in a zoological garden. A total of fourteen snakes from a similar source were examined. The isolations were made from a wide variety of the snake tissues. It appears quite significant that poults were dying on the majority of the ranches from which the snakes were collected. In many instances, the same Salmonella types were isolated from the poults and the snakes.

Hinshaw and McNeil (1944) examined three chicks that a garter snake had swallowed recently; the snake's liver was also examined. *S. meleagridis* and paracolon organism (type 8) were recovered from the liver of the snake. *S. panama* was recovered from the intestines of two of the chicks and from the pooled liver tissues of the three chicks. The same paracolon organism (type 8) was recovered from several other snakes that were captured near the turkey yards of the ranch. In an earlier study during the same season *S. meleagridis* had been isolated from several lots of poults that were being reared on the same ranch.

In another study, McNeil and Hinshaw (1944) investigated another outbreak of paratyphoid infection *(S. typhimurium)* among a brood of young poults on a certain ranch. The poults were hatched from breeding stock that was negative to both H and O types of *S. typhimurium* agglutination tests at the beginning of the hatching season. A retest of the flock after the outbreak occurred revealed a 3.7 per cent reaction among the breeding stock. Efforts were made to determine the source of infection. A garter snake, two quail, a crow, a king snake, two gopher snakes, three adult cats, and four young kittens were examined. *S. typhimurium* was isolated from two of the adult cats and from one of the garter snakes. The cats were known to be frequent visitors to the turkey yards. On a second ranch another outbreak occurred among the young poults. Again a live garter snake was captured in one of the brooder yards. Examination of the snake revealed the presence of *S. typhimurium* in the liver, spleen, testes, and intestines. Six weeks later, 320 house flies *(Musca domestica)* were caught on fly paper placed near two brooder houses on the second ranch. The flies were divided into eight lots of forty flies each and were examined. *S. typhimurium* was isolated from four of the lots. It is known that *S. enteritidis* can be transmitted through the complete life cycle of flies and that the infection may continue as long as four weeks within flies (Ostrolenk and Welch, 1942).

## PARATYPHOID IN TURKEYS

During the past two decades the turkey industry has been revolutionized. According to available records there were approximately ten million turkeys raised in 1890. Subsequently there was a gradual decrease so that during the period from 1910 to 1920 only about three million turkeys were raised annually. Since 1920 increasing numbers of turkeys were produced each year so that during the last few years there have been about thirty million birds raised annually. The marked decrease that occurred during 1910 to 1920 was mainly due to the losses resulting from enterohepatitis. Another important factor in the development of the turkey industry was the rapid growth made by the hatchery industry. Instead of depending upon a few breeding hens on the average farm and hatching the eggs under the turkey hens or chickens, the eggs are being incubated in forced draft incubators having a 50,000 to 60,000 egg capacity.

These developments in the industry have made it possible to incubate many millions of eggs each season. However, many new problems developed as the result of large-scale production. Other diseases have been recognized that caused losses among baby poults. Hewitt (1928) reported the isolation of S. *pullorum* from baby poults. This disease became widespread in a very short time. The rapid dissemination of the infection followed as the result of the large-scale production developed by the hatching industry. It has taken many years to develop means of controlling pullorum disease among the breeding turkeys. Unfortunately the turkey, especially the poult, is susceptible to many types of Salmonella other than S. *pullorum* and S. *gallinarum*, the two nonmotile types of the genus Salmonella. Paratyphoid infections are due to the types of Salmonella that are motile organisms. At the present time about forty types of paratyphoid organisms are known to have been responsible for losses in turkey poults. The adult turkey, which is the "carrier" of many types of Salmonella infection commonly encountered in the breeding stock, constitutes one of the major problems in the control of this disease. Deaths due to paratyphoid infection of adults are not of great economic importance.

Rettger, Plastridge, and Cameron (1933) reported that paratyphoid infection caused the death of many poults on two ranches situated about 90 miles apart. Turkeys had been raised for eleven years on farm A when, in the spring of 1929, the first evidence of an infection other than enterohepatitis appeared. At that time, about 25 per cent of a group of 200 young poults died. No turkeys had been brought to the farm during the past ten years, except 400 young poults in 1924 and a few adult toms which were purchased in 1931. In the spring of 1930, there were 1,000 breeding birds on the farm that produced 18,000 poults, of which 12,000 died during the hatching and brooding season. In 1931, 40 per cent of the poults died; during 1932, 2,600

poults were hatched, and 15 per cent of these died. The infection was intro-duced to farm B from farm A. The losses on farm B were quite comparable to those on farm A during the year of 1929, when the poults were obtained from farm A. A paratyphoid-like organism was isolated from the diseased poults. The strains recovered were considered as belonging to one species.

In 1934, Lee, Holm, and Murray (1936) encountered an acute disease that caused 90 per cent mortality among poults less than five weeks of age. In other flocks the losses were less, ranging from 40 to 70 per cent. A para-typhoid-like organism was isolated from the heart blood, liver, spleen, and bone marrow on liver-infusion agar and heart-infusion agar. The organism was found pathogenic for chicks, turkey poults, guinea pigs, and rabbits. The oral administration of 5 cc. doses of this paratyphoid organism to four turkey hens for 10 successive days resulted in the loss of weight and the development of diarrhea. Agglutination titers were found when the hens were tested sero-logically with antigens prepared from the cultures. Thirty days after expos-ure to the organism, the birds were destroyed and bacteriological examina-tions were made. The paratyphoid organism was recovered from three of the birds; the fourth was not examined. The organism was recovered from the ovaries of two of the birds. Other experimental turkeys developed an agglu-tination titer of 1:100 or above. Sixty eggs were produced and forty poults were hatched. During the first four weeks thirty of these poults died. The paratyphoid organism was recovered from twelve of the dead birds. The organism was isolated from the ovaries of four of the ten breeding hens which had produced the eggs. Only eight of the eighty-one eggs cultured could definitely be traced to the reactor hens.

Cherrington, Gildow, and Moore (1937) found an organism of the S. aertrycke type responsible for the loss of many poults on several turkey ranches in Idaho. The mortality was as high as 80 per cent during the first week of brooding. A group of twelve-week-old poults was purchased and put on a range with 300 of the older poults which had survived the disease. In approximately two weeks some of the added turkeys began to die. The organism involved in the original outbreak was recovered from the internal organs of the purchased turkeys. A group of the breeding stock was tested with antigen prepared from the organism involved, and more than one-third of the birds were found to have titers of 1:50 or higher. Twenty-two infertile eggs were examined bacteriologically, but the S. aertrycke type organism was not recovered. From three eggs out of a group of thirty which contained embryos dead in the shell, the S. aertrycke type organism was recovered. Five hens which had stopped laying but continued to have an agglutination titer were destroyed and examined bacteriologically. The paratyphoid organism was not recovered even though mishappen yolk sacs were present. During a second outbreak approximately one-third of the breeding flock reacted when tested with S. aertrycke antigen. Two dozen embryos, dead

in the shell, and a like number of infertile eggs from this flock failed to yield the causative organism. Five paratyphoid reactors from the flock showed a high percentage of abnormal egg yolks in the ovary, but the causative organism was not recovered. When another group of six infected hens (as determined by the agglutination test) was examined culturally, *S. aertrycke* type organisms were recovered from the ovaries of two birds. Thirty embryos, dead in the shell, were cultured from one flock; three of these yielded a *S. aertrycke* type organism. The results of these investigations indicate that the initial infection was present in the ovaries of the breeding hens, transmitted to fertile eggs or directly to the hatched poults, and possibly further disseminated in the incubator and brooder.

The records taken from unpublished data of the Diagnosis Laboratory, St. Paul, Minnesota (1927–46) are of considerable interest, indicating the increase in the incidence of paratyphoid infection in poults in recent years. During 1932, 4 poults; 1933, 10 poults; 1935, 14 poults; 1936, 45 poults; 1937, 45 poults; 1938, 39 poults; 1939, 120 poults; 1940, 117 poults; 1941, 103 poults; 1942, 137 poults; 1943, 48 poults; 1944, 52 poults; and 1945, 394 poults yielded an organism of *S. aertrycke* determined by bacteriological examination. Pomeroy and Fenstermacher (1939) first observed this infection in turkeys in 1932 when four poults were found affected with this disease. From 1932 to 1937 the *S. aertrycke* type of organism was recovered from poults which originated from thirty-one widely separated flocks. The disease was acute, with the losses varying from 10 to 25 per cent; in a few isolated cases the losses were as high as 90 per cent. The infection appeared in poults ranging in age from a few days to five weeks. On one farm the disease occurred in 1935 and reappeared the next year. The organisms isolated in 1935 were found pathogenic for guinea pigs, rabbits, and young poults. The breeding flock, consisting of 155 birds, was tested with whole-blood pullorum and an aertrycke plate antigen in March of 1937. About 8.5 per cent of the birds reacted and were removed from the flock. The disease did not appear during the 1937 season. Three other flocks in which the disease was known to have existed during the previous season were also tested. Eight per cent of the birds reacted and were removed from the flock. The disease did not appear during the season in which the breeding birds were tested. Entirely different results were obtained, however, in later studies.

During January, 1936, as the result of a request sent to a number of turkey breeders, 584 adult birds were tested by the whole-blood method, using pullorum and aertrycke antigens. Seven birds showed slight reactions to these tests and were purchased. This group was then tested by the tube method. Eight Salmonella antigens were used: *S. abortus equi, S. aertrycke, S. enteritidis, S. gallinarum, S. paratyphi A, S. paratyphi B, S. pullorum,* and *S. suipestifer.* The titers are not included but will be found with other details in the reference cited (Pomeroy and Fenstermacher, 1939). Three hens had

a maximum titer of 1:100 when tested with aertrycke and pullorum tube antigens. One hundred and three eggs were incubated, from which fourteen living poults were hatched. Seventy-eight eggs failed to hatch, eleven of which were broken; the latter were not examined, but the remaining eggs were either infertile, contained partially developed embryos, or were full-term embryos which died in the shell. Sixty-three eggs were found sterile; *S. aertrycke* type organisms were not recovered. The first poult died on the third day and two more died during the third week. The bacteriological examination gave negative results. The remaining eleven poults were raised to maturity, their blood sera tested at monthly intervals by the tube method, using the eight antigens listed previously. Negative results were obtained. When the hens were destroyed at the end of the hatching period the internal organs were examined bacteriologically. *S. aertrycke* and *S. pullorum* were not recovered.

The studies were continued during 1937. Four flocks containing 841 birds were available for study. Sixty-eight of these birds were found to have agglutinins present when tested by the whole-blood method, using aertrycke and pullorum plate antigens. Blood was collected from the sixty-eight birds and tested by the tube method. Only six of this group agglutinated aertrycke antigen in the 1:100 dilution; sera of thirteen others had a maximum titer of 1:50. Nine of these turkeys were purchased. A tom whose blood serum was negative when tested was added to the group. Three hundred and twenty-five eggs were incubated. Of this number, 90 were infertile by the end of the first week of incubation, 24 embryos were dead at the end of the second week of incubation, and 32 additional dead embryos were removed at the end of the third week of incubation. At the end of the fourth week, 57 more dead embryos were found. From the remaining 122 eggs, 115 living poults were hatched; the rest cracked the shells but failed to emerge. *S. aertrycke* was isolated from three infertile eggs, from one egg that contained a dead embryo at the end of 14 days of incubation, from two eggs that contained dead embryos at the end of 21 days of incubation, and from one egg that contained a dead poult. *E. coli* was recovered from 36 eggs, staphylococci from 27 eggs, Pseudomonas from 3 eggs, and Gram-positive rods from 15; 115 eggs were found sterile. Eighty-eight of the 115 poults died or were destroyed because they were crippled. *S. newington* was isolated from three and *S. montevideo* from one. The adults were destroyed and the internal organs subjected to careful bacteriological examination. *S. pullorum* was not re-covered, but *S. aertrycke* was recovered from the ovaries of three birds.

Three turkey hens were repeatedly fed a saline suspension containing viable *S. aertrycke* type organisms. Blood cultures were made, but *S. aertrycke* was not recovered. Agglutination titers appeared on the seventh day, reached a maximum point on the thirteenth day, and began to decline on

the twenty-first day. Living organisms were recovered from the feces on the day following the initial feeding. For four days following the last feeding of the organisms, positive results were obtained by fecal examination. Eggs from the above hens were collected and incubated. *S. aertrycke* was not isolated from infertile eggs or dead embryos. The hens were destroyed five months after being fed the cultures and the internal organs examined bacteriologically. Negative results were obtained.

Edwards identified the *S. aertrycke* type organisms which were isolated in this study and those recovered in the Diagnosis Laboratory. They were identified as follows: *S. aertrycke, S. anatum, S. bareilly, S. bredeney, S. derby, S. montevideo, S. newington,* and *S. senftenberg.*

Edwards (1937a) identified as *S. senftenberg* two cultures that had been isolated from young poults by Black. These cultures were isolated in 1936, and this constitutes the first recognition of this species in birds. The mortality did not exceed 10 per cent of the flock.

Edwards, Bruner, and Hinshaw (1940) isolated seven cultures of undescribed Salmonella type during an outbreak of a disease among young poults. Six of these cultures were isolated from a flock of poults assembled in California for the study of Hexamita infection. Intercurrent diseases were found in the flock. Three of the cultures were recovered from poults that originated from a single hatchery. The first deaths occurred when the poults were six days old. A mortality of 26 per cent occurred during the ensuing week. Three additional cultures were isolated from poults originating from different sources, but the poults had been placed in the experimental flock after being hatched. The seventh culture was obtained from a poult submitted for diagnosis. The seven cultures were found serologically identical and were named *S. california.* The *S. california* cultures were found to cross agglutinate with other types but were considered more closely related to *S. oranienburg.*

The isolation of two or more Salmonella species from one individual is not unusual. Edwards and Bruner (1940) have contributed extensively in this field of study. Many Salmonella types have been submitted to them for typing and more specific identification. In many instances two or more cultures were obtained from a single flock. Serological examination often indicates that more than one Salmonella type is involved in a single outbreak of paratyphoid infection. A number of types within the genus have been found to be transmitted through the egg. Edwards and Bruner (1940) reported twenty-six cultures from turkeys were found to contain twenty-five cultures of *S. typhimurium* and one of *S. derby.* Two cultures of *S. derby* and one each of *S. typhimurium* and *S. give* were isolated from another outbreak. Twelve cultures from another outbreak included eleven of *S. typhimurium* and one of *S. bredeney.* In another outbreak one culture examined con-

tained *S. anatum* and *S. derby*. In another culture isolated by Pomeroy, there were three types: *S. anatum, S. litchfield,* and *S. saint paul. S. litchfield* and *S. saint paul* were previously undescribed members of the Salmonella group. The culture originated from the liver of a single poult.

In a more comprehensive study Edwards (1939) examined 223 cultures of the Salmonella group. Of these, 54 had been isolated from chickens, 81 from turkeys, 22 from ducks, 60 from pigeons, 4 from canaries, 1 from a quail, and 1 from a pheasant. The following distribution of types was found:

| | | |
|---|---|---|
| *S. typhimurium* .....176 | *S. bareilly* .............4 | *S. montevideo* ........2 |
| *S. anatum* .............8 | *S. newport* ...........3 | *S. muenchen* ..........2 |
| *S. newington* . .......7 | *S. bredeney* ...........2 | *S. oranienburg* ........2 |
| *S. senftenburg* .. .. ...5 | *S. kentucky* .... .. . ..2 | *S. worthington* ........2 |
| *S. derby* ..............5 | *S. london* .............2 | *S. minnesota* .... .. .1 |
| | | *S. new brunswick* ......1 |

Pomeroy and Fenstermacher (1941) found that *S. typhimurium* organisms, when mixed with sterile turkey feces and then smeared on one-third of the outer surface of turkey eggs, were able to penetrate the egg shell during incubation and invade the egg contents. Eggs containing living embryos at the end of two weeks of incubation were inoculated in the air cell with .01 cc. of a saline suspension of *S. typhimurium*. Forty-five eggs were inoculated in this manner, and only four poults hatched. From fifteen control eggs inoculated similarly with .01 cc. of sterile saline, seven poults were hatched. A group of twenty-four eggs which were found infertile at the end of one week of incubation were inoculated in the air cell with 0.25 cc. of a 24-hour broth culture of *S. typhimurium*. The eggs were kept continuously in an incubator at a temperature maintained at 99.5° F. dry bulb and 83° and 84° F. wet bulb. Egg contents were examined bacteriologically at monthly intervals. Viable organisms were still being found at the end of thirteen months of continuous incubation. The experiment was discontinued at that point as the inoculated egg supply became exhausted. In another experiment 100 hatching turkey eggs were inoculated in the air cell with a saline suspension of *S. typhimurium*. From this group only 3 poults were hatched in comparison with 22 of the 50 saline-inoculated eggs and 28 of the 50 uninoculated control eggs.

Gauger and Greaves (1946) examined the egg contents of eggs laid by birds naturally infected with *S. typhimurium* and the shell and contents of eggs laid by birds experimentally infected with typhimurium organisms. *S. typhimurium* was not recovered from the contents of 164 eggs laid by six naturally infected "carrier" birds that were serologically positive and voided *S. typhimurium* in their feces quite regularly during the period the eggs were laid, including 34 eggs which were laid by one bird from which *S. typhimurium* was isolated from three infected ova. The experimentally infected turkeys became positive as indicated by serological test. At autopsy

eight of the original twelve turkeys were still positive to agglutination tests. The shell and egg content of 117 eggs laid by this group were examined. *S. typhimurium* was recovered from the shell of 27 eggs and 6 of the eggs yielded *S. typhimurium* from the shell and contents. Two of these 6 eggs had cracked shells at the time of examination. The egg content alone was not found to be infected in any case. Pomeroy and Fenstermacher (1941) incubated the eggs that were smeared with sterile feces mixed with *S. typhimurium* and found that the organisms pentrated the egg shell. There is urgent need for further study to determine to what extent paratyphoid organisms may penetrate the shell when laid by "carrier" birds.

Leg deformities in young and practically mature birds are not uncommon. Higgins, Christiansen, and Schroeder (1944) encountered a flock of twenty-four-week-old turkeys where 10 per cent of the birds were so severely affected that they were unsuitable for market. The condition was not similar to hock disorders observed during the latter part of the growing period. The affliction was of an inflammatory nature, showing evidence of swelling, heat, and interference with locomotion. *S. enteritidis* was isolated from the blood stream and from the tendon sheath as well.

Unpublished data of the Diagnosis Laboratory (1927–46) at St. Paul, Minnesota, indicate that day-old poults, when fed a suspension of *S. typhimurium*, usually die within a period of 10 days; the mortality may be 60 to 100 per cent. Adult turkey hens when fed viable organisms show very little evidence of enteric disturbance, but the organisms may become established in the digestive tract and be eliminated in the droppings.

The study of paratyphoid infection of turkeys has been a major project at the Diagnosis Laboratory for a number of years, and one of the many phases that has been investigated included the viability of paratyphoid organisms when subjected to experimental conditions as outlined by Pomeroy and Fenstermacher (1939). In this study turkey feces were sterilized, and then viable broth cultures of Salmonella were added to the sterilized feces. Turkey egg shells were sterilized and then smeared with the infected feces. The egg shells and feces were then placed in an incubator operated at 37.5° C. with 78 per cent humidity. Other egg shells and feces were placed in a covered box and stored in an animal building whose winter temperature was maintained near 50° F. Other egg shells were placed in another covered box and placed outside the above building and exposed to the elements. The following paratyphoids were included in the trials: *S. typhimurium, S. derby, S. montevideo, S. newington, S. bredeney, S. senftenberg, S. bareilly,* and *S. anatum.* The study was started in January and was continued for one year. It was found that all of the paratyphoid types smeared on the egg shells placed in the incubator were still viable at the end of eleven months. The viability of the organisms in the feces that were placed

in the incubator varied from 77 days to eleven months. The viability of the organisms on the egg shells placed in a box and stored in the animal building varied from 191 to 346 days. Only the egg shells were examined that were put in a box and placed outside the building and exposed to the elements. The viability of the organisms varied from 135 days to 350 days. Under the different experimental conditions there was no correlation of the period of viability for the different type of organisms. Therefore it is impossible to state that any single one of the types remained viable for the maximum period of time as stated.

In another experiment of a similar type, the broth cultures of the same paratyphoids were added to unsterilized turkey feces. The egg shells were placed in the incubator, and all were found to harbor viable organisms as long as 300 days. The viability of the organisms added to the feces and placed in the incubator varied from 17 to 24 days. The viability of the organisms on the shells placed in a box and stored in the animal building varied from 72 to 250 days. The viability varied from 28 to 38 days in feces under the same conditions. The viability of the organisms on the egg shells placed outside of the animal building varied from 54 to 156 days. Under similar conditions the viability in the feces varied from 11 to 156 days. In this experiment the inoculated egg shells and feces were also stored in a refrigerator maintained at 35° F. The viability of the organisms on the egg shells varied from 100 to 219 days. In feces stored under similar conditions the viability varied from 24 to 156 days.

Additional paratyphoid types are being reported constantly by research workers. Pomeroy (1944) listed 27 Salmonella types recovered from poults examined at the St. Paul laboratory. Since that time thirteen types have been added to the list, all of which have been identified by Edwards and Bruner. The following are the Salmonella types, not including *S. pullorum* and *S. gallinarum*, that have been isolated from turkeys in Minnesota:

| | | |
|---|---|---|
| *S. anatum* | *S. kentucky* | *S. paratyphi* B |
| *S. bareilly* | *S. lexington* | *S. rubislaw* |
| *S. bredeney* | *S. litchfield* | *S. saint paul* |
| *S. california* | *S. madelia* | *S. san diego* |
| *S. cerro* | *S. manhattan* | *S. senftenberg* |
| *S. chester* | *S. meleagridis* | *S. simsburg* |
| *S. cholera-suis* | *S. minnesota* | *S. tel aviv* |
| var. *kunzendorf* | *S. montevideo* | *S. thompson* |
| *S. derby* | *S. new brunswick* | *S. typhimurium* |
| *S. eastbourne* | *S. newington* | *S. typhimurium* |
| *S. enteritidis* | *S. newport* | var. *cophenhagen* |
| *S. gaminara* | *S. oranienburg* | *S. urbana* |
| *S. give* | *S. oregon* | *S. wichita* |
| *S. illinois* | *S. panama* | *S. worthington* |

Pomeroy (1944) reported on the mortality of young poults that had been fed regular mash for 2 and 4 days and then were fed a broth culture of *S. typhimurium*. The mortality of the poults infected when 2 days old varied from 40 to 60 per cent, and in those of the 4-day group the loss was 40 per cent. Other experimental evidence indicated that the older the poults are when exposed the lower is the expected mortality. Losses generally began 2 to 3 days after exposure and discontinued by the time the poults were two weeks old. In actual field cases the course of the disease is longer than would be expected under experimental conditions.

**Symptoms** in turkeys are not sufficiently characteristic to make a definite diagnosis. Droopiness, huddling near the heating element, ruffled feathers, and diarrhea may be observed. Intestinal disturbance, if present, may be nonspecific as far as paratyphoid infection is concerned. Unthriftiness may appear, especially if the disease continues through a prolonged course. Losses among young birds vary greatly depending upon the general sanitary conditions of the premises. Poults maintained in clean quarters may not suffer heavy losses. On the other hand the mortality may vary from 10 per cent to nearly 100 per cent. Symptoms have not been observed in adult turkeys.

Atypical outbreaks of this disease are confusing, especially in flocks of breeding turkeys which have been repeatedly retested with specific types of Salmonella isolated from dead poults. At times comparatively few losses during the early part of the hatching season are experienced, but later losses become serious in poults up to 10 days of age. The mortality may reach 60 to 80 per cent in the late hatches, and the paratyphoid organisms may be recovered readily from the infected poults.

**Lesions** may be entirely absent. Congestion of the liver and spleen are observed in some cases. Liver changes may include areas of focal necrosis. Lung and heart lesions are seldom present. Enteritis involving the duodenum is a common occurrence in poults that die from 3 to 5 days following artificial exposure to paratyphoid infection. Cecal cores are found quite frequently in experimental infections. These begin to form in birds that die in about 5 days after being infected; the longer they survive, the larger the cecal cores become. Adult birds rarely show evidence of the acute form of the disease, but they may become "carriers." The infection may localize in various organs, but few lesions develop. Adult "carrier" birds may void viable organisms in the feces for various periods without appreciable changes in the appearance of the intestinal mucosa.

**Diagnosis** should be established only by means of bacteriological examination with the isolation of the organism and its identification. Most laboratories are able to identify the organism in the Salmonella group and differentiate it from those of pullorum disease and fowl typhoid. Specific serotyping must be done in laboratories which are properly equipped for this purpose.

**Differential diagnosis:** Cecal cores have been mentioned as a post-mortem finding. Occasionally similar formations are found in the ceca of poults affected with pullorum disease. Careful search should be made to determine the presence or absence of lesions found quite commonly in pullorum infection which consist of nodules in the heart muscles and in the lungs. Their presence is strongly suggestive of pullorum disease. Pullorum disease and paratyphoid infection may be found in the same poult. Although this occurrence is not frequently reported it does occur often enough so that the possibility should not be overlooked. A number of cases have been found in which *S. pullorum* was isolated from the heart and liver, and one or more of the paratyphoids were recovered from the intestinal contents of a single poult. Veterinarians called to examine poults should make only a tentative diagnosis and inform the owner that the definite diagnosis can be made only by a laboratory equipped to make such an examination.

## REFERENCES

Altman, I. E.: 1940. *Salmonella suipestifer* infection in canaries. Jour. Am. Vet. Med. Assn. 97:601.

Beaudette, F. R.: 1926a. *B. aertrycke* infection in canary birds and parrots. Jour. Am. Vet. Med. Assn. 68:642.

———: 1926b. *B. aertrycke* as the etiological agent in a disease affecting squabs. Jour. Am. Vet. Med. Assn. 68:644.

——— and Edwards, P. R.: 1926. The etiology of a canary bird epizootic. Jour. Bact. 12:51.

Bruner, D. W., and Edwards, P. R.: 1941. Microorganisms of group E of the genus Salmonella with special reference to a new Salmonella type. Am. Jour. Hyg. 34 (Sec. B.) :82.

Cherrington, V. A., Gildow, E. M., and Moore, P.: 1937. Paratyphoid in turkeys. Poultry Sci. 16:226.

Clarenburg, A.: 1939. Paratyphoid in ducks in relation to public health. Proc. Seventh World's Poultry Cong. P. 233.

Cunningham, C. H.: 1941. Paratyphoid infection in quail. Jour. Am. Vet. Med. Assn. 99:217.

Darby, C. W., and Stafseth, H. J.: 1942. Salmonella infections common to man, animals, and birds. Proc. 46th Ann. Meet. U. S. Livestock Sanitary Assn. P. 189.

Doyle, T. M.: 1927. *B. aertrycke* infection of chicks. Jour. Comp. Path. and Therap. 40:71.

Edwards, P. R.: 1929. A fatal infection of chicks due to bacilli of the paratyphoid B group. Jour. Infect. Dis. 45:191.

———: 1935a. The antigens of *Salmonella anatum*. Jour. Bact. 30:269.

———: 1935b. A serological variant of *Salmonella aertrycke* isolated from pigeons. Jour. Bact. 30:465.

———: 1936. The occurrence of Salmonella, oranienburg type, in an infection of quail. Jour. Bact. 32:259.

———: 1937a. The occurrence of Salmonella, senftenberg type, in a disease in turkeys. Jour. Bact. 33:193.

———: 1937b. Paratyphoid infection of fowls. Jour. Am. Vet. Med. Assn. 90:403.

———: 1938. A new Salmonella type: *Salmonella kentucky*. Jour. Hyg. 38:306.

———: 1939. Incidence of Salmonella types in fowls in the United States. Proc. Seventh World's Poultry Cong. P. 271.

——— and Bruner, D. W.: 1938. Two new Salmonella types isolated from fowls. Jour. Hyg. 38:716.

——— and Bruner, D. W.: 1940. The occurrence of multiple types of paratyphoid bacilli in infections of fowls, with special reference to two new Salmonella species. Jour. Infect. Dis. 66:218.

——— and Bruner, D. W.: 1943. The occurrence and distribution of Salmonella types in the United States. Jour. Infect. Dis. 72:58.

———. Bruner, D. W., and Hinshaw, W. R.: 1940. A new Salmonella type isolated from turkeys: *Salmonella california*. Jour. Infect. Dis. 66:127.

——— and Rettger, L. F.: 1927. The paratyphoid B-suipestifer group of bacteria. Jour. Bact. 13:73.

Emmel, M. W., and Stafseth, H. J.: 1929. *Salmonella aertrycke* infection in the canary bird. Jour. Am Vet. Med. Assn. 75:230.

Gaiger, S. H., and Davies, G. O.: 1933. *Salmonella aertrycke* infection in chicks. Vet. Record 13:538.

Garside, J. S., and Gordon, R. F.: 1943–44. Salmonella infections of ducks and ducklings. Jour. Comp. Path. and Therap. 53:80.

Gauger, H. C., and Greaves, R. E.: 1946. Bacteriological examination of shells and contents of eggs laid by turkeys naturally or artificially infected with *Salmonella typhimurium*. Poultry Sci. 25:119.

———, Greaves, R. E., and Cook, F. W.: 1940. Paratyphoid of pigeons. I. Serological, bacteriological, and hematological studies of spontaneously infected birds. N. C. Agr. Exper. Sta., Tech. Bul. 62.

Gordon, R. F., and Buxton, A.: 1945. The isolation of *Salmonella thompson* from outbreaks of disease in chicks. Jour. Hyg. 44:179.

Graham, R.: 1936. Salmonella isolated from baby quail. Jour. Am. Vet. Med. Assn. 88:763.

Hewitt, E. A.: 1928. Bacillary white diarrhea in baby turkeys. Cornell Vet. 18:272.

Higgins, W. A., Christiansen, J. B., and Schroeder, C. H.: 1944. A *Salmonella enteritidis* infection associated with leg deformity in turkeys. Poultry Sci. 23:340.

Hinshaw, W. R., and McNeil, E.: 1944. Gopher snakes as carriers of salmonellosis and paracolon infections. Cornell Vet. 34:248.

——— and McNeil, E.: 1945. Salmonella types isolated from snakes. Am. Jour. Vet. Res. 6:264.

———, McNeil, E., and Taylor, T. J.: 1944. Avian salmonellosis. Am. Jour. Hyg. 40:264.

———, Taylor, T. J., and McNeil, E.: 1942. *Salmonella bredeney* infection in birds. Cornell Vet. 32:337.

Hoffman, H. A., and Edwards, P. R.: 1937. The spontaneous transmission of IV-variants of *Salmonella aertrycke* from pigeons to rabbits. Am. Jour. Hyg. 26:135.

———, Jones, E. E., and Stover, D. E.: 1943. Paratyphoid and paracolon infections in chickens and turkeys. St. Calif. Dept. of Agr. Bul., Vol. 32:66.

Hole, N.: 1932. Salmonella infections in ducklings. Jour. Comp. Path. and Therap. 45:161.

Hudson, C. B.: 1942. An outbreak of paratyphoid in guineas. Jour. Am. Vet. Med. Assn. 100:438.

Jungherr, E., and Clancy, C. F.: 1939. Serological types of Salmonella isolated from paratyphoid in chicks. Jour. Infect. Dis. 64:1.

——— and Wilcox, K. S.: 1934. *Salmonella aertrycke* as an etiologic agent of paratyphoid in pigeons. Jour. Infect. Dis. 55:390.

Khalifa, I. A. B.: 1935. Serological study of pigeon paratyphoid in Egypt. Jour. Am. Vet. Med. Assn. 86:24.

Lahaye, J., and Willems, R.: 1927. Une maladie des pigeons due à un germe du groupe des salmonella. Ann. de méd. vét. 72:241.

Lee, C. D., Holm, G., and Murray, C.: 1936. Paratyphoid infection in turkeys. Jour. Am. Vet. Med. Assn. 89:65.

Levine, N. D., and Graham, R.: 1942. Paratyphoid in baby wood ducks. Jour. Am. Vet. Med. Assn. 100:240.

Lütje, F.: 1921. Abort und Sterilität der Stuten. Deutsch. tierärztl. Wochenschr. 29:453.

Manninger, R.: 1913. Über eine durch den *Bazillus paratyphi B* verursachte Infektionskrankheit der Finken. Zentralbl. f. Bakt. I. Orig. 70:12.

———: 1918. Über Paratyphus beim Wassergeflügel. Allatorvosi Lapok, Budapest, p. 165. (Abst. in Jahresbr. Vet. Med. 38:160.)

McNeil, E., and Hinshaw, W. R.: 1944. Snakes, cats and flies as carriers of *Salmonella typhimurium*. Poultry Sci. 23:456.

——— and Hinshaw, W. R.: 1946. Salmonella from Galapagos turtles, a Gila monster and an iguana. Am. Jour. Vet. Res. 7:62.

Mohler, J. R.: 1904. Infectious enteritis of pigeons. Ann. Rep. of Bur. An. Ind., U.S.D.A. P. 29.

Moore, V. A.: 1895. On a pathogenic bacillus of the hog-cholera group associated with a fatal disease in pigeons. Bur. An. Ind., U.S.D.A., Bul. 8:71.

Morcos, Z.: 1935. Pigeon paratyphoid. Vet. Jour. 91:11.

Niemeyer, W. E.: 1939. Paratyphoid and trichomonas infection in pigeons. Jour. Am. Vet. Med. Assn. 94:434.

Ostrolenk, M., and Welch, H.: 1942. The house fly as a vector of food poisoning organisms in food producing establishments. Am. Jour. Pub. Health 32:487.

Pfeiler, W.: 1920. Beitrag zur Kasuistik des Hühnertyphus. Zeitschr. f. Fleisch- und Milch- Hyg. 30:267.

——— and Rehse, A.: 1913. Ueber das Vorkommen von Bakterien aus der Gruppe der Fleischchvergifter bei Vögeln. Paratyphus B-Infektion beim Huhn. Zentralbl. f. Bakt. I. Orig. 68:174.

Pomeroy, B. S.: 1944. Salmonellosis of turkeys. Doctoral thesis. Univ. Minn.

——— and Fenstermacher, R.: 1939. Paratyphoid infection of turkeys. Jour. Am. Vet. Med. Assn. 94:90.

——— and Fenstermacher, R.: 1941. Paratyphoid infection of turkeys. Am. Jour. Vet. Res. 2:285.

——— and Fenstermacher, R.: 1948. Sulfonamides in the control of salmonellosis of chicks and poults. Am. Jour. Vet. Res. (In press.)

Rettger, L. F., Plastridge, W. N., and Cameron, R.: 1933. Endemic paratyphoid infection in turkeys. Jour. Infect. Dis. 53:272.

Rettger, L. F., and Scoville, M.: 1920. *Bacterium anatum* n.s. the etiologic factor in a widespread disease of young ducklings known in some places as "keel." Jour. Infect. Dis. 26:217.

Schalm, O. W.: 1937. Study of a paratyphoid infection in chicks. Jour. Infect. Dis. 61:208.

Shirlaw, J. F., and Iyer, S. G.: 1937. A note on a variety of *S. enteritidis* isolated from pigeons. Indian Jour. Vet. Sci. and An. Husb. 7:231.

Spray, R. S., and Doyle, L. P.: 1921. Paratyphoid bacilli from chicks. Jour. Infect. Dis. 28:43.

Unpublished data from Diagnosis Laboratory, University Farm, St. Paul, Minn., 1927–46.

Van Roekel, H., and Bullis, K. L.: 1937. Salmonella infections in chickens. Jour. Am. Vet. Med. Assn. 91:48.

Weisgerber and Müller, C.: 1922. Untersuchungen über eine seuchenhafte Erkrankung der jungen Gänse in der Provinz Ostpreussen mit Paratyphus Befund. Deutsch. tierärztl. Wochenschr. 30:663.

# FOWL TYPHOID

By L. D. BUSHNELL, *Department of Bacteriology,*
*Kansas State College, Manhattan, Kansas*

☆　　☆　　☆

Fowl typhoid is a septicemic disease of domesticated birds. Its course is rather acute, and the mortality is high. However, its mortality, as a rule, is not as high and its course is not as rapid as that of fowl cholera or of fowl plague. It appears to be primarily a disease of chickens, but in exceptional cases ducks, turkeys, pheasants, peacocks, guineas, and a few other birds are attacked.

**History.** In 1888 a chicken breeder in England lost 400 chickens as a result of an infectious disease which was at first considered to be fowl cholera. Two hundred of these birds died in the first two months of the outbreak. Specimens were sent to Klein (1889) for autopsy and diagnosis. He reported it chiefly as an infectious enteritis. The intestinal mucosa and serosa were inflamed, and the feces appeared thin and greenish yellow. The spleen was enlarged two to three times; the liver was also somewhat enlarged, soft, flabby, and moist. The cause was an organism which he named *Bacillus gallinarum*. The same year he reported the disease among grouse, and in 1893 a similar disease among pheasants. The disease was investigated by Smith in Rhode Island in 1894 and more fully by Moore in Virginia and Maryland in 1895. Moore (1895) described the disease as "infectious leukemia" and named the organism *Bacillus sanguinarium*.

Klein observed small numbers of bacilli in the blood. They were non-motile, Gram-negative, and were easily cultivated. Chickens inoculated subcutaneously became sick in 5 to 6 days and died 2 to 3 days later. A similar disease was described by Lucet (1891) in France. Lignières and Zabala (1905) described a disease which was probably identical with Klein's disease. The catarrhal enteritis and the swollen spleen attracted attention; Gram-negative bacilli were observed in the blood. They were different from those described by Klein in that they first coagulated and later peptonized milk with an alkaline reaction.

Curtice (1902) studied the disease in Rhode Island and named it "fowl typhoid." The disease has been found in Germany, Hungary, Austria, France, Holland, and North and South America, as well as in Algiers. In

Germany it was observed by Pfeiler and Rehse (1913). Van Straaten and te Hennepe (1918) in Holland described the disease very fully.

On the basis of the post-mortem observation, Klein believed that it was not cholera, but a special disease. His suspicion was soon confirmed, because he ascertained that the newly discovered organism was different morphologically and biologically from that of fowl cholera.

**Transmission.** Like most other bacterial diseases fowl typhoid is spread in many ways. The source of most outbreaks is undoubtedly a recovered bird which has become a carrier and is discharging the organisms in such a manner as to contaminate the surroundings.

Attendants, feed dealers, chicken buyers, and visitors who travel from house to house and from farm to farm can carry the infection unless precautions are taken to disinfect footwear, hands, and clothing. Trucks, crates, and feed sacks are also very important. Wild birds, animals, and flies are important mechanical spreaders, especially if they have been feeding on carcasses of dead birds.

Egg transmission studies were reported by Simms (1946a). The fowl typhoid organism was isolated from infertile eggs, chick embryos, and baby chicks from survivors of both naturally and artificially infected hens. It was found that 37 per cent of the hens from the inoculated group and 27 per cent of birds recovering from a natural infection laid infected eggs. In the first group 10 per cent of the eggs laid were infected compared to 4 per cent in the second group. The feeding of these eggs caused heavy losses in susceptible birds. It is believed that the habit of egg eating by some hens may be an important source of fowl typhoid outbreaks when apparently healthy carriers are present. The effectiveness of the agglutination test at 30-day intervals, and the removal of reactors, however slight, was demonstrated in large commercial breeding flocks.

**Distribution.** The disease is widespread in the poultry-producing areas of the country, but outbreaks are sporadic, depending on factors which are not completely recognized at present. The distribution changes from year to year and season to season, although the seasonal variation seems to be more marked in the northern than in the southern parts of this country.

According to reports from Europe, it is now more widespread than before the first World War, although there has been a steady decline in incidence during the past ten years. The outbreaks vary widely from year to year and are considerably higher in the summer than in the winter, although Kaupp and Dearstyne (1925) report that the disease is most prevalent in North Carolina during the late winter and early spring, reaching its peak in April. Truche (1923), in France, and te Hennepe and van Straaten (1921), in Holland, report the disease as most prevalent in the spring and early summer. Apparently the seasonal prevalence is variable in different localities. The

reasons for its variation are not clear, although it has been suggested that it may be due to the fact that young adult birds are more susceptible than old birds or very young chicks. It has been suggested that it is a dry weather disease, but there it little evidence to support this statement.

According to Hall (1946), fowl typhoid has become much more prevalent in eastern United States during the past few years. It is especially severe in some broiler-raising areas. He reports that birds of all ages, from baby chicks to breeding hens, are affected, and that the disease occurs with equal frequency in young and mature stock. Losses range in different outbreaks from an occasional bird in old breeding flocks up to 75 per cent or more in younger fowls.

Glover and Henderson (1946) report an outbreak in chickens in Canada and state that it is believed to be the first case reported in that country.

**Etiology.** The causative agent of fowl typhoid is a relatively short, plump rod about 1.0 to 2.0μ long and 1.5μ in diameter. It has received the following names: *B. gallinarum, B. sanguinarium, B. typhi gallinarum alcalifaciens, B. paradysenteriae gallinarum, Eberthella sanguinaria, Shigella gallinarum,* and *Salmonella gallinarum.*

The bacilli mostly occur singly but are occasionally united in pairs. They have a tendency to stain a little heavier at the poles than in the center. They are Gram-negative, form no spores and no capsules, grow aerobically, and are nonmotile.

Gelatin colonies: Small, grayish-white, entire.

Gelatin stab: Slight, grayish-white surface; filiform growth in stab, no liquefaction.

Agar colonies: Moist, grayish, circular, entire.

Broth: Turbid with heavy flocculent sediment.

Litmus milk: Reaction unchanged, becoming translucent, no coagulation.

Indol: Not formed.

Nitrates: Reduced to nitrites.

Acid but no gas from dextrose, levulose, galactose, mannose, xylose, arabinose, maltose, dextrin, mannitol, dulcitol, and isodulcitol.

Hydrogen sulfide formed (depending on test used).

Facultative aerobe.

Optimum temperature 37° C.

The Duisburg variety differs from *Salmonella gallinarum* in its slow fermentation of maltose and in not forming $H_2S$. This organism is capable of undergoing rough variation followed by certain changes in its antigenic relations and sensitivity to bacteriophage action.

There has been considerable difficulty in the classification of the organisms of the fowl typhoid group due to their close relationship to *S. pullorum.*

Certain European investigators, including van Heelsbergen (1929), Manninger (1930), Miessner (1930), Wagener (1934), and Haupt (1935), have considered these organisms as identical species.

Smith (1915) and Smith and Ten Broeck (1915) were among the first to present a comparative study of *S. gallinarum* and *S. pullorum*. Their studies included agglutination relationships, toxin producing properties, and fermentation reactions. They noted the antigenic likeness of these organisms to each other and to those of *E. typhi*. Although the organisms were much alike, there were differences which caused them to be placed in different species. The basis for this decision was the production of gas by most strains of *S. pullorum* and the fermentation of maltose by *S. gallinarum*.

Goldberg (1917) reported that there was a difference in action in milk, dulcitol, isodulcitol, and dextrin.

Hadley *et al.* (1917) found that *S. pullorum* and *S. gallinarum* produced approximately the same amount of acid from glucose. However, maltose was fermented more rapidly and in greater amounts by *S. gallinarum*.

Mulsow (1919) first noted that certain strains of *S. pullorum* fermented maltose but suggested dulcitol as a better differential compound. This reaction has since been noted by others when serum-water is used as a basic medium. According to Edwards (1928) acid production in maltose by *S. pullorum* results from slow alkalinization of the medium during long incubations and from subsequent hydrolysis of this sugar by the alkali formed. Rodrigues and Pacheco (1936) confirmed these results and found in serum an enzyme that hydrolyzes maltose. They recommended caution in using serum-water as a basic medium for studying maltose fermentation.

Van Roekel (1935) reported a laboratory strain of *S. pullorum* which decomposed maltose after several years as a nonfermenting type for this sugar. He later (1937) isolated stable variants which subsequently fermented maltose within a few hours at 37° C.

Various differential media have been prepared for separating these two species. Cruickshank (1927) utilized Andrade's lactose agar for the original isolation from tissues. If growth of Gram-negative nonlactose-fermenting bacteria occurred in 12 hours at 37° C., an inoculation was made into a double agar medium containing 1.0 per cent maltose, 0.1 per cent glucose, and Andrade's indicator. *Salmonella gallinarum* produces a permanent deep red color in both the slant and the butt of the tube in 18 to 24 hours. *Salmonella pullorum* usually produces gas and a faint pink color in the butt of the tube, the slant remaining colorless.

Mallmann and Snyder (1929) used dextrin-lactose agar and dulcitol-lactose agar for differentiation. The media consisted of a 2.0 per cent basal agar containing 1.0 per cent dextrin or dulcitol and 0.5 per cent lactose with bromthymol blue as an indicator. With *S. gallinarum* the top was blue and

butt yellow; *S. pullorum* produced no change; *E. coli* produced a yellow-to-blue top and a yellow butt; and *P. avicida* a blue top and a green butt.

Pacheco (1935, 1936) and Pacheco and Rodrigues (1935) compared members of the *pullorum-gallinarum* group and some intermediate types. They used the neutral-red fluorescent test for gas production and a modified Drigalski medium, in which maltose was used, to separate the maltose from the nonmaltose fermenters. They also reported on the use of sorbite instead of lactose in modifying the medium. *S. pullorum* ferments sorbite but not maltose; *S. gallinarum* ferments maltose but not sorbite. See Table 1.

TABLE 1

ILLUSTRATING THE CLOSE RELATIONSHIP OF CERTAIN ATYPICAL *Salmonella pullorum*
AND *Salmonella gallinarum* STRAINS

|  | S. pullorum Gas-Forming Type | S. pullorum Non-Gas-Forming Type | Intermediate | S. gallinarum |
|---|---|---|---|---|
| Neutral red......... | fluorescent | — | — | — |
| Jordan's tartrate media........ ... | unchanged | unchanged | unchanged | yellow |
| Maltose.. ......... | — | — | A | A |
| Dulcite............. | — | — | A | A |
| Sorbite............. | A | A | A | — |
| Xylose............. | A | A | — | A |

— = no change; A = acid.

Table 1 summarizes Pacheco and Rodrigues' findings. It will be noted that the intermediates are similar to *S. gallinarum* in most respects. These writers recognize two types of *S. pullorum*, those producing gas as well as non-gas producers. Such characteristics are considered fairly constant for both types.

Hendrickson (1927) recognized that *S. gallinarum* was maltose-dextrin-dulcite positive, while *S. pullorum* fails to ferment these compounds. He seemed to agree with Hadley *et al.* (1917) that *Bact. pullorum* may be *Bact. sanguinarium* in the making.

Monteverde and Simeone (1944) reported that *S. gallinarum* cultures isolated in Argentina regularly fermented maltose and dulcitol with production of acid, but never attacked sorbitol. The reaction on *d*-tartrate was irregular. The production of $H_2S$ was inconsistent.

Brown, Duncan, and Henry (1924) reported on the use of organic acids for differentiating these species. They used these in 1.0 per cent concentration in a basic medium with phenol-red as an indicator. Twenty different species of Salmonella, including *S. gallinarum* and *S. pullorum*, were studied. The salts of formic, citric, and *d*-tartaric acids were most useful but were not entirely satisfactory. Because of irregular results, this method was considered less reliable than the fermentation methods using sugars.

Mallmann (1931b) tested several salts of organic acids. Of these, mucic and *d*-tartaric had merit and gave very consistent results. *S. pullorum* gave an alkaline and *S. gallinarum* an acid reaction. Hinshaw and Rettger (1936) confirmed these observations. The *Salmonella jeffersonii* gave reactions identical to those of *Salmonella gallinarum*. This may indicate that these organisms are the same as suggested by St. Johns-Brooks and Rhodes (1923).

In regard to the production of $H_2S$ by these organisms, there is considerable difference of opinion, some of which is probably due to different methods of testing. According to Truche (1923) the reaction is stronger with *S. pullorum* than with *S. gallinarum,* and Klimmer and Haupt (1927) state that the reverse is true. Pacheco and Rodrigues studied the reactions on a variety of media with lead acetate, bismuth with and without cysteine, iron salts, and a peptone gelatin. *S. pullorum* produced $H_2S$ rapidly on agar with lead and bismuth, both with and without cysteine, and slowly on the other media.

Hinshaw (1941) was able to separate the two species by use of a 0.15 per cent cysteine hydrochloride gelatin medium. *Salmonella gallinarum* in 89 of 91 strains produced a characteristic yellowish-white or grayish turbidity when incubated at 37° C. for 72 hours. *S. pullorum* did not produce such changes. Several maltose-fermenting variants of *S. pullorum* have been observed among those studied. None of these gave a positive reaction either in the cysteine-gelatin medium or in tartrate agar. He concludes that although many variants exist, there is increasing evidence that these organisms are a distinct species.

Hinshaw reports that $H_2S$ can be demonstrated readily in cultures of *S. gallinarum* growing in cyteine-gelatin by its faint odor and by use of strips of lead acetate paper. Only a faint browning of such paper is noted in *S. pullorum* cultures grown in this medium.

Table 2 contains a summary of the characteristics of variants of *S. pullorum* and *S. gallinarum*.

It is stated that there are probably several subspecies in the *pullorum-gallinarum* group. The Van Roekel maltose-fermenting strain gave reactions identical with those of the California maltose-fermenting strains. These strains differ from the *Salmonella intermedius* A-type strains in that the latter are xylose-negative and dulcite-positive. The *S. intermedius* B-types were sensitive to *S. pullorum* bacteriophage and were considered by Nobrega (1935) as variants of the latter organism. Hinshaw (1941) found that *S. intermedius* A and B types reacted like *S. pullorum* in both cysteine-gelatin and tartrate agar. He states that the maltose-negative strain of *S. gallinarum* of Van Roekel was so classified on the ground that it produced a positive *S. gallinarum* reaction in cysteine-gelatin, acid in tartrate-agar, and was dulcitol-positive.

The dulcitol-negative *S. gallinarum* of Kujiumgieff differs from that of Delpy and Rastegar's (1938) type B, in that the latter is dulcitol-positive and tartrate-negative.

The Duisburg strains of *S. gallinarum* are like *S. pullorum* in that they do not ferment tartrate agar and do not give any reaction in cysteine-gelatin. They resemble *S. gallinarum* in the type of growth on nutrient agar, and in

TABLE 2

SUMMARY OF CHARACTERISTICS OF VARIANTS OF *Salmonella pullorum* AND *Salmonella gallinarum* (HINSHAW, 1941)

Fermentation Reactions

| Variants | Maltose | Xylose | Dulcite | Arabinose | Cysteine-Gelatin | Tartrate Agar (J-H) |
|---|---|---|---|---|---|---|
| S. pullorum (Van Roekel)........ | AG* | AG | — | AG | — | — |
| S. pullorum (9 Calif. cult.)...... | AG* | AG | — | ‡ or AG | — | — |
| S. intermedius A type (4 cult.)......... | ‡ or AG | — | AG | AG | — | — |
| S. intermedius B type (2 cult.)......... | A | — | A | A | — | — |
| S. gallinarum (Kujiumgieff)........ | A | — | — | A | — | A |
| S. gallinarum (Barboni)........... | — | A | A | — | — | A |
| S. gallinarum (Van Roekel)........ | — | A | A | A | T | A |
| S. gallinarum (Duisburg) (2 cult.)... | A | ‡ or A | A | A | — | — |

AG = acid and gas; A = acid; ‡ = variable or slow reaction; T = yellowish-white or grayish turbidity in media.

\* No maltose-fermenting strains were isolated in Kansas.

being maltose and dulcitol positive. They are more nearly like the *S. intermedius* type B.

Johnson and Rettger (1942) studied the basal nutrition of *S. gallinarum* and *S. pullorum* with sixteen amino acids and thioglycolic acid. With the exception of two strains which required nicotinic acid or its amide, forty-three strains of *S. pullorum* did not require any vitamins, while vitamin $B_1$ proved to be highly indispensable for the growth of twenty-two strains of *S. gallinarum*. Nearly all strains of *S. pullorum* required glucose while *S. gallinarum* did not. None of the strains of the former required the addition of $CO_2$, while several of the latter required an appreciable amount for growth. Leucine was the most important single amino acid; tryptophane was not required.

**Serological relationships.** Although there are certain cultural and physiological differences between *S. gallinarum* and *S. pullorum,* their serological and antigenic characters are identical.

Beck and Eber, Beaudette, Hadley *et al.,* Hendrickson, Mulsow, May, Goodner, Rettger, Koser, Manninger, van Heelsbergen, and others have tried to separate the *S. gallinarum* and the *S. pullorum* on the basis of the agglutination reactions but have not succeeded. Even by applying the absorption method, the two organisms proved to be identical. Kauffmann (1930, 1934) states that *S. gallinarum* and *S. pullorum* strains contain the same O-antigen. This has been confirmed by Edwards (1939) in more recent studies. Many investigators consider that it is not improbable that *S. gallinarum* and *S. pullorum* represent two varieties of one microorganism. (See particular description under pullorum disease.)

Rodrigues and Pacheco (1936) were unable to detect any antigenic difference between *S. gallinarum* and the intermediate types. Although some appear to doubt that it is worthwhile to differentiate between the organism of fowl typhoid and pullorum disease, most American investigators agree that the organisms are somewhat different in their fermentation reactions and the pathological changes which they produce in infected birds. The agglutination test will not differentiate between carriers of either type of infection. Hinshaw (1941) was able to differentiate the two organisms by the use of a 0.15 per cent cysteine-hydrochloride gelatin medium first reported by Hinshaw and Rettger (1936). After incubation at a temperature that does not liquefy the gelatin, a turbid halo appeared around the individual colonies in shake cultures, and along the line of inoculation in stab cultures. None of the 454 strains of *S. pullorum* consistently produced visible change in the medium, or at best a surface pellicle. Species of bacteria which were irregular in their reactions, but which gave mostly negative results were fourteen strains of *E. typhi* and three strains of *S. anatum.* A few cultures of Pseudomonas and Proteus from turkeys gave these reactions.

The Jordan-Harmon sodium-potassium-tartrate medium (1928) was a valuable supplementary medium to use with the cysteine-gelatin. The *S. gallinarum* strains consistently produced acid on this medium while *S. pullorum* produced no change.

**Resistance.** In general the resistance of this organism is about the same as that of the other members of the typhoid and paratyphoid groups.

The fowl typhoid organism is killed within 10 minutes at 60° C. It remained viable in the dark for 20 days in ordinary and in distilled water, but dies in 24 hours when exposed to sunlight. When dried on glass plates and kept in the dark, the organism retains its viability for 89 hours; under the action of direct sunlight it is killed in a few minutes. The organism is killed by phenol in a 1:1,000 dilution and by bichloride of mercury in a 1:20,000

dilution, potassium permanganate 1.0 per cent in 3 minutes, and 2.0 per cent formalin in 1 minute. Agar cultures rapidly lose their pathogenic character, although they retain their antigenic properties for some time. According to Altara, *S. gallinarum* can be demonstrated in the bone marrow of carcasses in the virulent state three months after chickens have died of fowl typhoid. No doubt under certain conditions it lives for much longer periods. Kaupp and Dearstyne (1924) reported that although direct sunlight destroyed the organism in a short time, it remained viable for 20 days when stored in water in the dark.

The resistance of this organism to the action of bacteriophage is of some interest. D'Herelle (1919–22) examined the excreta of fowls and tested the bacteriophage for virulence against eight strains of bacteria. Bacteriophage activity was demonstrated from all excreta studied; some samples showed marked activity for all cultures used. This investigator claimed that his experiments with bacteriophage confirmed his conclusion that the immunity to an infection is assured at a time when the body contains a bacteriophage virulent for that organism. Mallmann (1931a) criticizes these observations because of lack of controls. It was found that the bacteriophage from one organism was easily adapted to another by use of mixed cultures. Bacteriophage was of no value in treating chicks either naturally or artificially infected. Munné (1937) obtained several cultures of *S. gallinarum* and *S. pullorum*, all of which were equally susceptible to the action of bacteriophage.

**Pathogenicity.** The pathogenicity of fowl typhoid cultures has proved decidedly variable in experiments with chickens. Infection by mouth was not always successful. Likewise, fowls cannot always be fatally infected subcutaneously. Intramuscular injection is the most effective method of infection. Kaupp and Dearstyne (1925) reported sixteen deaths out of forty chickens artificially infected; fifteen became visibly sick, and nine showed no symptoms. Similar results have been obtained at this station, whereas others report from 25 to 90 per cent loss.

Epidemiologically there are a few peculiarities in regard to the disease. Van Heelsbergen (1929) states that according to his experience it is very difficult, in some cases at least, to infect chickens which come from a region to which fowl typhoid is indigenous. If chickens are imported from a part of the country where the disease is not known, infection is rather easy. It is suggested that the bacteriophage, or acquired immunity, is probably in part responsible.

Although this is primarily a disease of young adults, fowl typhoid has been reported in young birds by several investigators. Beaudette (1925), Beach and Davis (1927), Martinaglia (1929), Komarov (1932), Hinshaw and Taylor (1933), having isolated the organism from the ovary of a turkey, suggested a possible transmission through the egg. St. Johns-Brooks and

Rhodes (1923) found that strains of the *S. gallinarum* produced lesions in young chicks indistinguishable from those associated with pullorum disease.

A relatively small number of avian species appear to be susceptible. Lucet (1891) described what was probably an outbreak of the disease in turkeys but claimed that ducks, geese, and pigeons were not susceptible. Donatien *et al.* (1923) consider palmipeds to be refractory, but found the turkey, guinea fowl, and pea fowl among the susceptible species; ducks and geese were resistant. Pfeiler and Roepke (1917) mention the pheasant, turkey, and guinea fowl as susceptible in natural outbreaks, but that ducks, geese, and pigeons are not, although a duck which had been inoculated with a culture died a few days later. Kaupp and Dearstyne (1924), Beck and Eber (1929), and te Hennepe (1924) have observed the disease in ducks. Kaupp and Dearstyne (1925) state that turkeys are less susceptible than chickens, and that guineas, though slightly susceptible, yield to artificial inoculation. Fox (1923) isolated *B. sanguinarium* from an outbreak of disease among parrots in The Philadelphia Zoological Garden. Beck and Eber (1929) reported on the loss in ducklings 1 to 14 days old due to *B. gallinarum* infection. Truche (1923) found that pheasants, swans, grouse, sparrows, ring doves, and ostriches commonly became infected, but that the duck, goose, and turkey were more resistant. Johnson and Anderson (1933) reported outbreaks of the disease in ducklings, turkeys, and guinea fowl. The infection has been observed in wild birds, in quail, grouse, and pheasants. These birds are susceptible by feeding or injection of cultures. Te Hennepe (1939) states that fowl typhoid has decreased in the Netherlands during the past ten years from a point at which it caused some 8.0 per cent of the total deaths in adult birds to 0.7 per cent in 1939. This is considered to be due to greater interest in poultry diseases and improved care of poultry. The disease was at one time one of the most important in Kansas. About 1935 it practically disappeared and is still quite uncommon. The reason for this is not known. El-Dine (1939), in Egypt, states that fowl typhoid is often mistaken for fowl cholera. He reports the disease mainly in chickens and turkeys, and states that it has been reported in peacocks but has never been seen in pigeons, geese, or ducks. A vaccine was used to confer immunity.

The reports on the susceptibility of pigeons have been variable. Klein reported no success following subcutaneous injection of cultures. Lucet (1891) was unable to infect pigeons with 1.0 cc. doses subcutaneously, while Moore (1895) killed pigeons within 8 days with 2.0 cc. of a broth culture. Pfeiler and Roepke (1917) killed pigeons by injecting 1.0 cc. of a 24-hour broth culture, but the heart blood of these birds would not cause infection in a second pigeon. Kaupp and Dearstyne (1924) caused the pigeon to become sick on the third or fourth day with recovery on the fifteenth day. Kraus (1918) produced death in a pigeon within 4 days by use of 1.0 cc. of

a 24-hour broth culture of the fowl typhoid organism. Te Hennepe and van Straaten (1921) claim that pigeons are not always susceptible to inoculation with these organisms.

Hinshaw (1930) reported that the disease caused greater losses among California turkeys than did blackhead. Hinshaw and Taylor (1933) reported an ovarian infection of a turkey hen from which they isolated the organism. Hudson and Beaudette (1929) state that there is a difference of opinion as to the incidence of fowl typhoid among avian species, but that there is evidence that it is greater than is ordinarily suspected. A lack of accurate field diagnosis undoubtedly results in many discrepancies. Lerche (1939) reports one case of an infection of humans with a culture very similar, but not identical, with *S. gallinarum*, and the Duisburg strain of *S. gallinarum* was originally obtained from acute gastroenteritis in man. Cloud (1943) reported the isolation of the Duisburg strain from a patient with severe peritonitis. The organism was not found in the stools. However, this organism should not be considered a human pathogen.

It is not difficult to infect rabbits with fowl typhoid bacilli. Pfeiler (1920) succeeded in inducing infection four times in fifteen rabbits. Guinea pigs and pigeons are very resistant, although, as has been observed repeatedly, pigeons die if the dosage is somewhat large. Van Straaten, te Hennepe, and Pfeiler were quite successful in infecting white and gray mice, while rats, dogs, and cats were shown to be immune. A relatively small number of avian species appear to be susceptible although there is a difference of opinion as to the incidence of this disease. Hinshaw and Taylor (1933) inoculated two mature rabbits intravenously with 0.5 and 1.0 cc., respectively, of a 48-hour broth culture; the animals lived. The one receiving 1.0 cc. was killed three weeks later, and the blood was found infectious for young rabbits. Th. Smith and Ten Broeck (1915)· stated that the pathogenicity for *S. gallinarum* was relatively feeble for laboratory animals. The rabbit succumbed to relatively large doses (0.3 to 0.5 cc.) of a 24-hour broth culture given intravenously. Pfeiler and Rehse (1913) state that pigeons, geese, and ducks are resistant to the infection, but mice succumb. Rats, cats, and dogs fail to show any disturbance after eating diseased material.

Smith and Ten Broeck found a toxin in the filtrates of broth cultures of *S. gallinarum*. It appeared in the culture at the end of 2 days at 37° C. and caused prompt death of a rabbit by the intravenous route. Death resulted within 2 hours and in many respects was like an anaphylactic shock. It is probably an endotoxin which is stable at 60° C. for 1 hour. Boiling for 15 minutes reduces its activity.

**Symptoms.** In the North Central United States the peak of the seasonal losses from fowl typhoid is approximately four months later than that of pullorum disease. In this area the former is chiefly a disease of young adults

and is rare in chicks. In some parts of the world this disease causes most deaths in spring and at the beginning of summer, at the same time that pullorum disease prevails. The incubation period, as a rule, is from 4 to 5 days, although this varies considerably with the virulence of the organism, and the course of the disease is about 5 days. The disease may be considered a septicemia accompanied by a well-marked toxemia. The injury to the intestinal mucosa may open the way for the rapid absorption of nonspecific toxic agents from the intestine. The injury to the liver causes an increased destruction of erythrocytes, as indicated by the anemia and the marked increase in bile pigments in the intestine, resulting in a greenish diarrhea. The birds become listless and inactive and prefer to separate from the flock. A thin greenish-yellow diarrhea appears early; there is complete loss of appetite; intense thirst as in fowl cholera is common, presumably as the result of high fever. Investigations at the Kansas Station would indicate that there is no difference in the temperature range of fowl cholera and fowl typhoid; temperatures of 110° to 112° F. are common. Respiration is at first accelerated. In some cases death occurs suddenly at the end of the second day. Comb and wattles appear anemic instead of cyanotic as in fowl cholera. The anemic condition becomes pronounced in prolonged cases. Mortality is variable; there may be three to four deaths daily in a fairly good-sized flock.

In the cases observed by Pfeiler and Rehse, there were twelve deaths the first week and nineteen later on in a flock of eighty-seven chickens. All birds displayed a sickly appearance, indicating that they were probably all infected and suffered more or less from the infection. Fowls visibly sick generally do not survive.

In a flock of adult chickens suffering from chronic infection, the mortality as a whole is not so great. In such a flock there is an occasional death. The post-mortem picture indicates a chronic disease (foci in different organs, chronic fibrinous peritonitis). Some European investigators are of the opinion that chronic S. *pullorum* infection and fowl typhoid are two forms of the same disease. This opinion is not generally held in the United States. Rettger and Koser (1917) concluded as early as 1917 that despite the several characters which these organisms have in common, and particularly the serological reactions, that the organisms constitute two distinct types, and each holds a specific relationship to the disease with which it has been associated in the past.

**Pathology.** Rigor mortis occurs very rapidly. Comb and wattles are generally pale (in fowl cholera comb and wattles generally become darker). Van Straaten and te Hennepe point out, however, that the comb may also be dark colored in fowl typhoid. This has been observed at this laboratory when birds die soon after the beginning of the infection. Because of the diarrhea

the feathers around the vent are generally stuck together with greenish-yellow feces. Emaciation is not observed because of the short duration of the disease.

The first change is the extensive swelling of the spleen. Its normal weight is about 0.1 per cent; with fowl typhoid it may be as high as 0.3 per cent of the body weight. The liver is also markedly swollen, congested, soft in consistency, and shows fatty degeneration. Frequently very small, gray, necrotic foci are found beneath the capsule. The color of the liver varies from yellow to greenish-brown or bronze (mahogany colored). The weight of the liver normally is about 1.5 per cent; with fowl typhoid it may go up to 5.5 per cent of the body weight. The gall bladder is usually distended with thickened bile. An intestinal catarrh of varied intensity occurs. The exudate is either slimy and very abundant or more purulent and slight in amount. The exudate is characteristically greenish and bile-stained throughout. In some instances the catarrhal inflammation is limited to a few areas; in other cases it extends from the gizzard to the cloaca but ordinarily is most evident on the anterior third of the intestinal mucous membrane. The ceca are frequently very much inflamed. Furthermore, petechiae may occur in the mucous membrane. According to American reports the hemorrhagic character of the enteritis seems to be pronounced. The heart is sometimes enlarged, and frequently there is a severe pericarditis. Very common lesions observed in this laboratory are the small, irregular, grayish, necrotic areas on the liver and heart muscle. As a rule the kidneys are swollen and may exhibit areas of focal necrosis, but no marked changes are noticed in the lungs. In some instances there is slight edema and a few necrotic areas, but there is no severe congestion as in fowl cholera.

In acute cases the blood picture is changed very little. If the disease lasts a few days, however, the changes mentioned in Table 3 are observed.

The organism of fowl typhoid was formerly considered as causing an acute disease only of mature birds. However, in recent years it has been isolated

TABLE 3
CHANGES OF THE BLOOD WITH FOWL TYPHOID (WARD AND GALLAGHER, 1920)

| Date | Temperature in C. | Number of Red Blood Corpuscles per cm. | Number of White Blood Corpuscles per cm. | |
|------|-------------------|----------------------------------------|------------------------------------------|---|
| March 26.... | 41.5 | 3,535,000 | 18,940 | Healthy |
| March 28.... | 43.5 | 2,430,000 | 70,000 | Chicken eats very little |
| April 2....... | 43.8 | 1,684,210 | 80,000 | Blood very pale; chicken weak, refuses food |
| April 3....... | 41.3 | 1,745,000 | 245,000 | Very weak, very many red blood corpuscles attacked by leucocytes |
| April 4....... | .... | ........ | ...... | Found dead |

from dead chicks by several investigators. Hall (1946) reports heavy losses among young birds in battery brooders in eastern United States.

Beaudette (1925) isolated from the unabsorbed yolk of chicks and the ovaries of adult hens an organism which had the same fermentative power as the fowl typhoid bacillus. He believes that the infection may result from the contact between mature birds and carriers of the infection and may be passed through the egg to young chicks. Beach and Davis (1927) made similar observations.

Johnson and Pollard (1940) studied an outbreak of disease which resembled pullorum disease in week-old turkey poults. The autopsy revealed a large retained yolk, and the liver appeared somewhat friable and of creamy-white color. The surface was mottled with slight hemorrhagic areas. A slight congestion of the anterior portion of the duodenum was found. The organism isolated was a Gram-negative rod producing acid but no gas from dextrose, mannite, dulcite, xylose, sorbite, arabinose, maltose, levulose, and dextrin, but did not react with lactose, sucrose, or inositol. Serologically the organisms checked with *S. gallinarum*. This organism was isolated from the ovaries of the adult birds that supplied the poults. It appeared to be a very chronic disease in these birds. By testing at frequent intervals with a pullorum antigen the percentage of reactors in the flock was reduced from 8.7 to 6.0 per cent. However, it was considered as doubtful if the infection could be completely eliminated in this manner.

Beck and Eber (1929) recognized great loss from *B. gallinarum* infection among ducklings 1 to 14 days old. Maltose, dulcite, and dextrin were fermented with acid formation by the organism isolated. The disease picture was similar to the one observed in pullorum infection in chicks. The ducklings were sick only a short time. The anatomical changes were the following: hemorrhage in the pericardium, slight swelling of the spleen, catarrhal inflammation of the lungs and intestines. Small necrotic foci in the lungs, as frequently observed in chicks with pullorum disease, did not occur. In adult ducks the changes of the ovary and the yolk were frequently the same as those found in adult hens. Fowl typhoid organisms were isolated from the misshapen yolks.

Beaudette (1938) states that the disease in the guinea is interesting because the affected birds show respiratory symptoms characterized by a severe congestion with collection of mucus in the nasal cleft and trachea. The lungs were congested, and the organism could be isolated from the nasal exudate.

Gauger (1934) obtained the organism of fowl typhoid from focal lesions in the testicles of a rooster. The culture was pathogenic for other roosters by inoculation and feeding. Although various foci had been described, this was the first case described for focalization in the testicles.

**Diagnosis.** In general it is not difficult to diagnose fowl typhoid clinically.

The disease is not as acute as fowl cholera, monocytosis (pullet disease), or fowl plague. In these latter diseases, as many birds are sometimes lost in a few hours as are lost with fowl typhoid in as many days. On autopsy the differences exhibited by these diseases are generally quite evident. The marked swelling of the liver in fowl typhoid is not found in fowl cholera and fowl plague, and the general hemorrhagic character of the last two diseases is more common. Petechiae are observed in fowl cholera or fowl plague more frequently than in fowl typhoid. The bronze-colored liver makes almost certain the diagnosis of fowl typhoid. Also, the microscopic blood picture differs in fowl typhoid from that in fowl cholera and fowl plague. A few rods resembling colon organisms may be seen in the preparations from cases of fowl typhoid; in fowl cholera the bipolar organisms are very common; in plague no bacilli are found in the blood. As a means of differential diagnosis a chicken and a rabbit may be inoculated with some material from the diseased bird. In fowl cholera both animals die in 1 or 2 days; in plague, only the chicken dies in 2 to 4 days; in fowl typhoid, the rabbit generally survives, the chicken remains alive or dies after 6 to 10 days.

A laboratory diagnosis is quite easy. The organism grows readily on ordinary laboratory media and may be identified by bacteriological or serological methods. The following table will be of value in this diagnosis.  ·

TABLE 4

FERMENTATION REACTIONS OF COMMON POULTRY PATHOGENS

| | Dextrose | Lactose | Maltose | Sucrose | H₂S | Indol |
|---|---|---|---|---|---|---|
| *Sal. gallinarum*... . ... | A | — | A | — | ? | — |
| *Sal. pullorum*..... .. .. | AG | — | — * | — | + | — |
| *Sal. anatis*.............. | AG | — | AG | — | + | — |
| *Sal. typhimurium*..... . | AG | — | AG | — | + | — |
| *Past. avicida*..... | A | — | — | A | + | + |
| *E. coli*........... .. | AG | AG | AG | ? | — | + |
| *Staphylococci*....... .. | A | A | A | A | ‡ | — |

AG = acid and gas; A = acid; — = no change; ‡ = slight; + = positive.
* Of the large number of cultures of *S. pullorum* which were isolated in the Laboratory of Bacteriology, Manhattan, Kansas, the maltose-fermenting type was very rare.

The *S. anatis* may be separated from *S. typhimurium* by fermentation of inositol. *S. anatis* fails to ferment it while the latter ferments it with production of acid and gas.

A positive antipullorum serum will be of value in making a slide agglutination test. This, however, will not differentiate the two organisms, but is a valuable adjunct to the fermentation reactions.

Eosin-methylene blue, SS, bismuth sulfite, or desoxycholate citrate agar plates are suitable for the isolation of this group of organisms either directly or following enrichment. A small bit of liver tissue is rubbed over the entire

surface of the agar plates. After 24 hours at 37° C., a vigorous culture is usually obtained. This may be differentiated from the colon, aerogenes, and pullorum organisms by the appearance of the colonies which are of medium size and colorless. They will have the same appearance as those of the paratyphoids and some of the "slow lactose fermenters" of the colon group, and must be finally identified by means of $H_2S$ and indol production.

Mallmann *et al.* (1928) reported on the use of brilliant-green in the isolation of *S. gallinarum.* The growth of the organism was not affected by freshly prepared solutions of 1 to 75,000, while the dye exerted an inhibitory effect on *E. coli,* as previously described by others. The bacteriostatic effect of different lots of dye from different manufacturers was found to be decidedly different. These results were confirmed by Kerr (1930). Delpy and Rastegar (1938) found that this dye could be used to advantage for the isolation of the pullorum-gallinarum group and its intermediates.

Fowl typhoid in chickens may be confused with fowl cholera and monocytosis in the field. This is especially true if the diseases are acute. If the birds die quickly with fowl typhoid the liver is lighter than normal, with pale streaks. In monocytosis the most characteristic change of the liver consists of round, yellowish areas with hemorrhagic centers. No degeneration of the skeletal muscle is observed in fowl typhoid while it is fairly diagnostic for monocytosis.

It should be emphasized that the final differential diagnosis must be made in the laboratory where the organism can be isolated and identified (no organism can be isolated which will cause monocytosis).

The agglutination test, using antigen made from *S. gallinarum* or *S. pullorum,* can be used to detect the birds infected with this organism.

<div align="center">CONTROL</div>

**Serum.** It is quite certain that passive immunity can be obtained with highly potent immune serum. Such passive immunity, as a rule, does not last longer than two or three weeks. After this time the chickens have again reached their usual state of susceptibility. Since the infectious material may still be present, either in some carrier or in the soil, the birds are exposed to infection after losing the passive immunity so that in many cases the disease breaks out anew two to three weeks after the serum treatment. However, the use of such serum will probably never be an important means of controlling this disease because of the cost.

**Vaccines.** Attempts have been made to immunize fowl with killed fowl typhoid cultures. According to some writers the results of this type of treatment are of little value. However, it cannot be denied that killed bacteria of the group possess some antigenic properties. McNutt (1926) experimented with various commercial vaccines on about 725 chickens and con-

cluded that they have no value for the control of fowl typhoid. It is probable that repeated vaccination will produce a better degree of immunity than one treatment. Experiences from field practice point in that direction. Bushnell and Patton (1924) found that three vaccinations at about 5-day intervals reduced the mortality from some 30 to 5.6 per cent in birds occupying contaminated runs. The results of vaccination are difficult to evaluate because of the natural variation in normal resistance. Van Es and Olney (1940) exposed fowls to artificial infection and observed that the losses were variable and that the birds which contracted the disease showed a considerable degree of variation in the length of their survival. This difference was probably due to individual variation in susceptibility.

Simms (1946b) reported experiments on the use of bacterial vaccines in fowl typhoid. Vaccines were prepared by killing cultures of S. gallinarum with formalin, phenol, chloroform, brilliant-green, and crystal violet. All except the crystal violet vaccines were suspended in beeswax and peanut oil. Injections were made subcutaneously, intraperitoneally, intramuscularly, and into the crop. No significant protection was observed after the vaccinated birds had been exposed to infection later.

**Other methods.** Sanitation is by far the most important means of controlling any infectious disease of poultry. That is especially true of fowl typhoid. Van Es and Olney (1940) exposed birds to various degrees of sanitation and noted the effect on losses from fowl typhoid. Of the 80 birds which died, 10 were in the more sanitary and 70 in the less sanitary pens. The conclusion that sanitary measures are of value in the control of this disease is warranted.

Very sick birds should be killed and burned or buried deeply. The building must be thoroughly cleaned; walls, perches, nests, and apparatus must be scrubbed with lye water (1.0 pound of lye dissolved in 15 gallons of hot water). The upper surface of earth floors should be replaced with fresh soil; cement floors should be cleaned and disinfected daily during an outbreak. The dropping boards should be cleaned each day. Feeding pans and drinking vessels as well as all other utensils should be thoroughly scoured with lye water and rinsed. Sometimes a hot solution is preferable because of the resistance of protozoan parasites to the action of chemical disinfectants. This is especially true in case of coccidia and worm eggs. After the building has been cleaned, dried, and aired, the walls may be whitewashed. An antiseptic whitewash may be prepared by adding 5.0 per cent crude carbolic acid. The runs must be spaded at frequent intervals. Yards and runs should be arranged so that all parts are exposed to direct sunlight at some time during the day.

Give drinking water to which potassium permanganate (one teaspoon to 5 gallons of water) or one of the hypochlorites (0.5 parts of available chlor-

ine per million) has been added. As soon as the permanganate loses its purple color it must be replaced. The containers must be placed so that droppings and other organic material cannot get in; otherwise, both potassium permanganate and chlorine compounds immediately lose their disinfecting power due to the oxidation of noninfectious material. Puddles and pools must be eliminated. Pigeons, mice, and rats are to be kept away from the premises. If there are several chicken houses to be served by the same attendant, a mat dipped in disinfectant may be placed so that shoes are cleaned before entering each house.

Breeding and selection for resistant strains of birds may be an important means of control. Lambert (1933) showed that selection for resistance to fowl typhoid in chickens resulted in a decided decrease in the mortality of selected stocks. Since *S. gallinarum* and *S. pullorum* are closely related organisms, it was decided to test fowl typhoid resistant stock for susceptibility to *S. pullorum* infection, and an *S. pullorum* stock for susceptibility to *S. gallinarum*. Results indicated that selection for resistance to one pathogen affords some protection to infection with one closely related. It was suggested that the resistance was to some extent due to nonspecific factors.

Sulfonamide drugs have been tried by numerous investigators with conflicting results. Hammond (1945) reported effective control by use of sulfathiazole. Holtman and Fisher (1946) studied an outbreak in battery-raised chickens which had caused 20 per cent loss in 3 days. The flock was then divided into two parts. Group 1 received the usual care, removal of sick birds and cleaning and disinfection of batteries with a cresol solution. Group 2 was given sodium sulfathiazole in the drinking water for one week. At the end of the week 80 per cent of the birds in group 1 had died. Losses in group 2 had been reduced to 4 per cent with no losses during the last 3 days. However, losses in this group reappeared within 5 days but were controlled by the use of the drug. Simms (1946b) reported on the use of sulfamerazine, sulfadiazine, and sodium sulfathiazole in broiler plants. The drugs were fed in 0.5 to 1.0 per cent in wet mash. None of these drugs was satisfactory for controlling a virulent outbreak of the disease. Mortality was greatly reduced while the drugs were being fed, particularly in case of sulfamerazine, but on discontinuance of its use, mortality rose again to nearly its former level. The same results were obtained from the use of these drugs in breeding flocks.

Moore (1946) reported that sulfamerazine was effective in reducing death losses from fowl typhoid, while sulfathalidine and sulfasuxidine were not effective. The mortality varied from 33.3 to 83.3 per cent for these two drugs. However, the former was given 5 days after exposure while the latter was started at the time of exposure.

Jones, Metzger, Schatz, and Waksman (1944) reported on the use of

streptomycin to protect chick embryos from the action of the fowl typhoid organism.

## REFERENCES

Beach, J. R., and Davis, D. E.: 1927. Acute infection of chicks and chronic infection of the ovaries of hens caused by the fowl-typhoid organism. Hilgardia 2:411.

Beaudette, F. R.: 1925. The possible transmission of fowl typhoid through the egg. Jour. Am. Vet. Med. Assn. 67:741.

————: 1938. An outbreak of fowl typhoid in guineas. Jour. Am. Vet. Med. Assn. 92:695.

Beck, A., and Eber, R.: 1929. Die wichtigsten bakteriellen Kückenerkrankungen. Ihre Diagnose, Differentialdiagnose und Bekämpfung. Zeitschr. f. Infeckt.-Krankh. d. Haustiere 35:76.

Brown, H. C., Duncan, J. T., and Henry, T. A.: 1924. The fermentation of salts of organic acids as an aid to the differentiation of bacterial types. Jour. Hyg. (London) 23:1.

Bushnell, L. D., and Patton, J. W.: 1924. The use of vaccines in poultry diseases. Poultry Sci. 4:64.

Cloud, O. E.: 1943. Perforation with peritonitis from *Shigella gallinarum* (var. Duisburg). Med. Bul. Veterans' Admin. 19:335.

Cruickshank, G. A.: 1927. Employment of a double sugar medium for routine diagnosis of bacillary white diarrhea, fowl typhoid, and fowl cholera. Jour. Bact. 14:135.

Curtice, C.: 1902. Fowl Typhoid. R. I. Agr. Exper. Sta., Bul. 87.

Delpy, L., and Rastegar, R.: 1938. Étude de souches, américaines, asiatiques et éuropéennes de microbes du groupe pullorum-gallinarum. Ann. de l'Inst. Pasteur 61:536.

d'Herelle, F.: 1919. Sur le rôle du microbe bactériophage dans la typhose aviaire. Compt. rend. Acad. Sci. 169:932.

————: 1922. The Bacteriophage; Its Role in Immunity. 206–207. The Williams and Wilkins Co., Baltimore.

Donatien, A., Plantureaux, E., and Lestoquard, F.: 1923. La typhose aviare en Algérie. Ann. de l'Inst. Pasteur d'Algérie 1:585.

Edwards, P. R.: 1928. The fermentation of maltose by *Bacterium pullorum*. Jour. Bact. 15:235.

————: 1939. Standard strains of Salmonella. Ky. Agr. Exper. Sta., Circular 50.

El-Dine, H. S.: 1939. Important diseases of poultry in Egypt and their control. Proc Seventh World's Poultry Cong., p. 229.

Fox, H.: 1923. Diseases of Captive Wild Animals and Birds. First Edition. Lippincott Co., Philadelphia. P. 598.

Gauger, H. C.: 1934. A chronic carrier of fowl typhoid with testicular focalization. Jour. Am. Vet. Med. Assn. 84:248.

Glover, J. A., and Henderson, W.: 1946. Fowl typhoid. Report on a recent outbreak in Ontario. Jour. Comp. Med. 10:241.

Goldberg, S. A.: 1917. A study of the fermenting properties of *B. pullorum* (Rettger) and *B. sanguinarium* (Moore). Jour. Am. Vet. Med. Assn. 51:203.

Hadley, P., Caldwell, D. W., Elkins, M. W., and Lambert, D. J.: 1917. Infections caused by *Bacterium pullorum* in adult fowls. R. I. Agr. Exper. Sta., Bul. 172.

Hall, W. J.: 1946. Fowl typhoid. Cir. Vol. 755. U.S.D.A., pp. 1–9.

Hammond, J. C.: 1945. Sulfonamides in the control of fowl typhoid. Poultry Sci. 24:382–84.

Haupt, H.: 1935. IV. Zur Systematik der Bakterien. Die für Mensch und Tier pathogenen gram-negativen alkalibildenden Stäbchenbakterien. Ergeb. Hyg. Bakt. Immunit. u. Exper. Therap. 17:175.

Hendrickson, J. M.: 1927. The differentiation of *Bacterium pullorum* (Rettger) and *Bacterium sanguinarium* (Moore). Jour. Am. Vet. Med. Assn. 70:629.

Hinshaw, W. R.: 1930. Fowl typhoid of turkeys. Vet. Med. 25:514.

————: 1941. Cysteine and related compounds for differentiating members of the genus Salmonella. Hilgardia 13:583.

———— and Rettger, L. F.: 1936. Cysteine-gelatin as a differential medium for *Salmonella pullorum* and *Salmonella gallinarum*. Proc. Soc. Exper. Biol. and Med. 35:44.

———— and Taylor, T. J.: 1933. A chronic carrier of fowl typhoid of turkeys. Jour. Am. Vet. Med. Assn. 82:922.

Holtman, D. F., and Fisher, G.: 1946. Some observations on the control of fowl typhoid infection with sulfa drugs. Jour. Bact. 51:401.

Hudson, C. B., and Beaudette, F. R.: 1929. The isolation of *Bacterium pullorum* from a European bullfinch (*Pyrrhula europa*). Jour. Am. Vet. Med. Assn. 74:929.

Johnson, E. A., and Rettger, L. F.: 1942. A comparative study of the nutritional requirements of *Salmonella pullorum*, *Salmonella gallinarum*, and *Salmonella typhosa*. Jour. Bact. 43:103.

Johnson, E. P., and Anderson, G. W.: 1933. An outbreak of fowl typhoid in guinea fowls. (*Numida meleagris*). Jour. Am. Vet. Med. Assn. 82:258.

—— and Pollard M.: 1940. Fowl typhoid in turkey poults. Jour. Am. Vet. Med. Assn. 96:243.

Jones, D., Metzger, H. J., Schatz, A., and Waksman, S. A.: 1944. Control of gram-negative bacteria in experimental animals by streptomycin. Science 100:103.

Jordan, E. O., and Harmon, P. H.: 1928. A new differential medium for the paratyphoid group. Jour. Infect. Dis. 42:238.

Kauffmann, F.: 1930. Die Technik der Typenbestimmung in der Typhus-Paratyphus Gruppe. Zentralbl. f. Bakt. I. Orig. 119:152.

——: 1934. Untersuchungen über die Duisburger Gallinarum Stämme. *Ibid*. 132:337.

Kaupp, B. F., and Dearstyne, R. S.: 1924. Chronic carriers of fowl typhoid. Jour. Am. Vet. Med. Assn. 64:329.

—— and Dearstyne, R. S.: 1924. Fowl typhoid. A comparison of various European strains with those of North America. Poultry Sci. 3:119.

—— and Dearstyne, R. S.: 1925. Fowl typhoid and fowl cholera. N. C. Agr. Exper. Sta., Tech. Bul. 27.

—— and Dearstyne, R. S.: 1925. The differential diagnosis of fowl cholera and fowl typhoid. Jour. Am. Vet. Med. Assn. 67:249.

Kerr, W. R.: 1930. Selective media for the cultivation of *Bacillus pullorum* and *B. sanguinarium*. Jour. Comp. Path. and Therap. 43:77.

Klein, E.: 1889. Ueber eine epidemische Krankheit der Hühner, verursacht durch einen Bacillus— *Bacillus gallinarum*. Zentralbl. f. Bakt. 5:689.

Klimmer, M., and Haupt, H.: 1927. Ueber Infektion von Hühnern mit dem *Bacterium gallinarum* Klein (1889). Zentralbl. f. Bakt. I. Orig. 105:99.

Komarov, A.: 1932. Fowl typhoid in baby chicks. Vet. Record 12:1455.

Kraus, E. J.: 1918. Zur Kenntnis des Hühnertyphus. Zentralbl. f. Bakt. I. Orig. 82:282.

Lambert, W. V.: 1933. A preliminary study of the reaction of two disease resistant stocks of chickens after infection with their reciprocal pathogens. Iowa Acad. Sci. 40:231.

Lerche, M.: 1939. Salmonellainfektionen beim Geflügel und ihre Bedeutung für die Epidemiologie der Salmonellabakterien. Proc. Seventh World's Poultry Cong., p. 274.

Lignières, J., and Zabala: 1905. Sur une nouvelle maladie des poules. Bul. Soc. Cent. de Méd. Vét. 59:453.

Lucet, A.: 1891. Dysenterie épizootique des poules et des dindes. Ann. de l'Inst. Pasteur. 5:312.

McNutt, S. H.: 1926. Vaccination of poultry. Jour. Am. Vet. Med. Assn. 69:472.

Mallmann, W. L.: 1931a. Studies on bacteriophage in relation to salmonella and pullorum disease. Mich. Agr. Exper. Sta., Bul. 109.

——: 1931b. Use of organic acids for the differentiation of *Salmonella pullorum* and *Salmonella gallinarum*. Proc. Soc. Exper. Biol. and Med. 28:501.

—— and Snyder, D.: 1929. Differential medium for *Salmonella pullorum*, *Salmonella gallinarum*, *Pasteurella avicida*, and *Escherichia coli*. Jour. Infect. Dis. 44:13.

——, Thorp, F., and Semmer, M.: 1928. A medium for the isolation of *Salmonella pullorum* and other members of the paratyphoid group from avian tissues. Jour. Am. Vet. Med. Assn. 73:825.

Manninger, R.: 1930. Hühner typhus und bakterielle Kückenruhr. Proc. Eleventh Internat. Vet. Cong. (London). Part 3:724.

Martinaglia, G.: 1929. A note on *Salmonella gallinarum* infection of ten-day old chicks and adult turkeys. Jour. So. Africa Vet. Med. Assn. 1:35.

Miessner, H.: 1930. Die Pullorum Infektion der Hühner (Die weisse Ruhr des Kücken-Hühnertyphus). Deutsch. tierärztl. Wochenschr. 38:517.

Monteverde, J. J., and Simeone, D. N.: 1944. Salmonelas genuinamente avairias en aves "reaction antes." Univ. Buenos Aires Fac. Agron. y Vet. Inst. etc. 1–30 (Biol. Abs. Vol. 19. 1945).

Moore, E. N.: 1946. The efficacy of recently developed sulfonamides against fowl typhoid. Poultry Sci. 25:307.

Moore, V. A.: 1895. Infectious leukemia in fowls—A bacterial disease frequently mistaken for fowl cholera. U.S.D.A. Bur. An. Ind., 12th and 13th Ann. Rep.

Mulsow, F. W.: 1919. The differentiation and distribution of the paratyphoid-enteritidis group. IV. Avian paratyphoid bacilli: A comparative study of *B. pullorum* and *B. sanguinarium*. Jour. Infect. Dis. 25:135.

Munné, J. V.: 1937. Au sujet de la différentiation de *Salmonella pullorum* et *S. sanguinarium* au moyen d'un bactériophage spécifique. Compt. Rend. Soc. de Biol. 126:1228.

Nobrega, P.: 1935. Differenciacao entre "S. pullorum" e "S. gallinarum." Papel importante do bacteriophago. Arch. do Inst. Biol.—São Paulo 6:71.

Pacheco, G.: 1935. Biologie des bacteries du groupe pullorum-gallinarum action sur le milieu au lait et sur le rouge neutre. *Ibid*. 118:888.

——: 1936. Biologie du groupe pullorum-gallinarum. Caractérization des types qui composent le groupe. *Ibid*. 121:590.

—— and Rodrigues, C.: 1935. Nouveau representant des bacteries du groupe pullorum-gallinarum. Morphologie des colonies du groupe. Compt. Rend. Soc. de Biol. 118:905.

Pfeiler, W.: 1920. Identitätsnachweis für die Erreger der Kleinschen Hühnerseuche und des Pfeiler-Rehseschen-Hühner typhus Bazillus. Zentralbl. f. Bakt. I. Orig. 85:193.

—— and Rehse, A.: 1913. *Bacillus typhi gallinarum alcalifaciens* und die durch ihn verursachte Hühner seuche. Mitt, a.d. Kaiser Wilhelm Institut f. Landwirtschaft zu Bromberg. 5:306.

—— and Roepke, W.: 1917. Zweite Mitteilung über das Auftreten des Hühnertyphus und die Eigenschaften seines Erregers. Zentralbl. f. Bakt. I. Orig. 79:125.

Rettger, L. F., and Koser, S. A.: 1917. A comparative study of *Bacterium pullorum* (Rettger) and *Bacterium sanguinarium* (Moore). Jour. Med. Res. 35:443.

Rodrigues, C., and Pacheco, G.: 1936. Biologie du groupe pullorum-gallinarum. Observations relative à la differentiation des types pullorum par la fermentation de la maltose. Compt. Rend. Soc. de Biol. 123:438.

St. Johns-Brooks, R., and Rhodes, M.: 1923. The organism of the fowl typhoid group. Jour. Path. and Bact. 26:433.

Simms, B. T.: 1946a. Egg eating may spread fowl typhoid. Rep. Chief Bur. An. Ind. Agr. Res. Adm. U.S.D.A., p. 39.

——: 1946b. Tests of drugs and vaccine to control fowl typhoid. *Ibid*., p. 40.

Smith, Th.: 1915. A note on the relation between *B. pullorum* (Rettger) and the fowl typhoid bacillus (Moore). Jour. Med. Res. 31:547.

—— and Ten Broeck, C.: 1915. Agglutination affinities of a pathogenic bacillus from fowls (fowl typhoid) (*Bact. sanguinarium*, Moore) with the typhoid bacillus of man. Jour. Med. Res. 31:503.

te Hennepe, B. J. C.: 1924. Combating poultry diseases by the state serum institute at Rotterdam. Proc. Second World's Poultry Cong., p. 219.

——: 1939. Combating poultry diseases in the Netherlands. Proc. Seventh World's Poultry Cong., p. 224.

—— and van Straaten, H.: 1921. Fowl septicemia. Trans. First World's Poultry Cong. 1:259.

Truche, C.: 1923. De la typhose aviaire. Ann. de l'Inst. Pasteur 37:478.

Van Es, L., and Olney, J. F.: 1940. An inquiry into the influence of environment on the incidence of poultry diseases; Fowl typhus. Univ. of Neb., Res. Bul. 118.

van Heelsbergen, T.: 1929. Handbuch der Geflügelkrankheiten and Geflügelzucht. Ferdinand Enke, Stuttgart. P. 135.

Van Roekel, H.: 1935. A study of the variation of *Salmonella pullorum*. Mass. Agr. Exper. Sta., Bul. 319.

——: 1937. Maltose-fermenting *S. pullorum* strains. Mass. Agr. Exper. Sta., Bul. 339.

van Straaten, H., and te Hennepe, B. J. C.: 1918. Die Kleinsche Hühnerseuche. Folia Microbiol. 5:103.

Wagener, K.: 1934. Kückenruhr. Proc. Twelfth Internat. Vet. Cong. (New York) Part 3:108.

Ward, A. R., and Gallagher, B. A.: 1920. Diseases of Domesticated Birds. Macmillan Co., New York.

CHAPTER ELEVEN

## FOWL CHOLERA

*By* CHAS. MURRAY, *Dean Emeritus, Division of Veterinary Medicine,*
*Iowa State College, Ames, Iowa*

☆  ☆  ☆

Fowl cholera is a contagious disease affecting practically all classes of fowls, usually of a septicemic nature, characterized by petechial hemorrhages of the mucous membranes, and manifesting both high morbidity and mortality.

**History.** During the eighteenth century various epidemics among birds were designated as anthrax, typhus, or pest. Maillet, in 1836, cited by Manninger (1929), was the first to use the designation *fowl cholera* in connection with severe losses. However, it was not until the middle of the nineteenth century that the infectious nature of the disease was first recognized. Renault and Delafond, according to Manninger (1929), presented experimental evidence of the transmissibility of fowl cholera.

Rivolta, in 1877, and Perroncito, in 1878, described the presence of non-motile "coccobacilli" and diplococci in the blood of affected birds. In the absence of a bacteriologic identification, it is possible that these workers were dealing with other pathogenic forms (Hadley and Amison, 1911; Hadley, 1912). Pasteur (1880a) succeeded in growing pure cultures of the microorganism in neutral chicken broth. These studies scientifically established fowl cholera as a distinct disease entity and differentiated it from other infections.

Following these studies Pasteur (1880b), using this organism, performed his fundamental experiments in the attenuation of bacteria in culture and their use as immunizing agents.

The later work of Kitt (1888) included studies on the nature of fowl cholera as well as immunological investigations. Salmon (1880, 1881, 1883) was the first investigator to report on the presence of fowl cholera in the United States.

**Incidence.** Fowl cholera is widely distributed in most of the temperate and warm regions, less common in northern countries. It may occur as an enzootic in any of these, or merely sporadically. In some regions, at times, it may cause a heavy mortality, at others the losses may be nominal.

In Europe it is reported as having decidedly decreased in the past twenty

years, until it is of no special importance in northern, western, and central
Europe. In eastern and southern countries it is still prevalent. In mid-
western United States it is far less frequently reported than it was ten years
ago. In one Iowa diagnostic laboratory the receipt of positive specimens is
one-fifth what it was at that time.

In the United States the disease is more or less seasonal, most common
in wet or cold weather, uncommon in dry. Hoffman and Stover (1942), in a
study of 30,000 autopsies of chickens in California, found a close correlation
between number of cases of fowl cholera and amount of rainfall. It seldom
is seen in brooder chicks, even in those running in a flock in which it is
present in the adult fowls.

**Etiology.** *Pasteurella avicida* (*P. cholerae gallinarum, P. aviseptica,* etc.),
the causative agent of fowl cholera, is a small oval rod of the hemorrhagic
septicemia group. It is 0.25 to 0.4μ by 0.6 to 2.5μ in size, attaining the greater
length after repeated culture (Fig. 11.1). In general the microorganism is
pleomorphic, producing thread formation when cultivated on alkaline agar
or on carbohydrate bouillon. In recently isolated cultures from the acute
type of the disease, a capsule may be demonstrated, of a mucoid nature and
varying size, which disappears on prolonged cultivation.

In tissue or blood stained with methylene blue, Giemsa, and carbol-
fuchsin, the organism is distinctly bipolar, but only to a slight degree or not at
all from culture media. It is an aerobe and facultative anaerobe, with an opti-
mum growth temperature of 37° C. The pH growth range is from 6 to 8.5
with optimum 7.2 to 7.4 (Merchant, 1946).

Beef infusion medium is fairly favorable for growth, but is greatly im-
proved if blood serum is added. Some strains fail to grow except in media
containing blood serum. In broth the recently isolated culture produces
diffuse clouding. Some strains produce a flocculent precipitate, character-
istic of the rough phase of the organism. Aged broth cultures develop a
pellicle on the surface and a sticky sediment at the bottom of the tube.

Hughes (1930), in a study of 210 freshly isolated strains of *P. avicida,*
reports three types of the organism as distinguished on the basis of colony
morphology. One type, "fluorescent," is said to be associated with epidemics
of fowl cholera, is highly virulent, and so stable in suspension that it is
agglutinable only in very acid buffers (pH 2.4) and not at all in antisera.
The second type, "blue," occurs in flocks in which cholera is endemic,
possesses a low virulence, is unstable in suspension, agglutinating in acid
buffers over a wide zone (pH 2.4–5.6) and in all *P. avicida* antisera. The
third type, "intermediate," is associated with the more severe cholera and is
intermediate in behavior.

In fermentation studies as a basis for classification, Rosenbusch and Mer-
chant (1939) found that the typical hemorrhagic septicemia Pasteurellae

fall into three groups on the basis of their fermentation of xylose, arabinose, and dulcitol. Group I ferments arabinose and dulcitol but not xylose; Group II ferments xylose but not arabinose and dulcitol, while Group III is variable, more nearly like Group I. The five strains of *P. avicida* studied by them fell into Group I, four of which showed fluorescence corresponding to Hughes' first type. All five strains fermented glucose, levulose, galactose, and mannose, with acid production but no gas. Contrary to Hughes' experience, Rosenbusch found the strains he studied highly agglutinable, all five falling into the same class.

**Tenacity.** The bacteria of fowl cholera are preserved at least a month in manure (Gärtner, 1898), about three months in the decaying carcass and in garden soil (Kitt), and 18 days in water with the exclusion of air and at a temperature from —6° C. to —8° C. They also offer considerable resistance to cold (Kitt, 1888); in ice they are virulent at least 14 days. According to Hertel (1904) the virulence gradually weakens at —13° C. They become avirulent by drying in exudate in the open air with the reaction of sunlight within 48 hours, and with the exclusion of light within 72 hours. Organs of carcasses are sterilized at a temperature of 45° to 50° C. in three-fourths hour, but within 5 to 10 minutes at a temperature from 80° to 85° C. (Kitt, 1888).



Fig. 11.1. *Pasteurella multocida*, blood smear, fowl cholera. ×2,000. (From Nowak, Documenta Microbiologica, Gustav Fischer.)

Van Es and Olney (1940), in the course of experiments carried on at the University of Nebraska to determine the influence of environment on the incidence of fowl cholera, found that in a poultry yard in which, during a period of approximately three years, the disease had maintained itself, the infection hazard had apparently entirely disappeared two weeks after the yard had been vacated and after the occurrence of the last death due to the disease.

The following disinfectants react energetically: 1 per cent phenol, 3 per cent cresol, one-half per cent sulfuric acid, and 1–5,000 bicholoride of mercury. Blood and diarrheal discharges are safely disinfected by thoroughly mixing with 5 per cent copper sulfate solution.

**Susceptibility.** Domestic fowls of every type, as well as small feral birds (sparrows, finches, etc.) that visit chicken yards in order to find their food, are attacked by fowl cholera. Sparrows may spread the infection. Birds of

prey kept in zoological gardens likewise succumb to food infection. Chickens, pigeons, and waterfowl may be infected per os with more or less difficulty. Kaupp and Dearstyne (1925) state that only seventeen birds became sick and ten died in a flock of thirty-five that was fed virulent material.

Rabbits are very easily infected fatally by feeding and by inoculation with fowl cholera organisms. House and field mice are likewise susceptible. A subcutaneous inoculation with *P. avicida* produces only a local abscess in the guinea pig which, however, may succumb to intraperitoneal inoculation.

Horses, cattle, sheep, pigs, dogs, and cats are refractory to the infection in the digestive tract, but abscesses result after subcutaneous inoculation. All these animals succumb, however, to intravenous inoculation with moderately heavy doses. While man may generally consume without harm fowls that are suffering from the disease, it is advised that their meat should under no circumstances be used as human food.

**Sources of infection.** The infection is given off by diseased birds, especially in the body wastes which contaminate the soil as well as the food and water, which constitute perhaps the most important factor in the dissemination of the disease. The carcasses of fowls dead of cholera and thoroughly permeated by the organisms may serve as a source of infection for some time. These constitute a particular hazard because of the tendency of the healthy birds of the flock to consume such material. The most prolific source of infection for waterfowl is the contaminated water of pools, ponds, and ditches in which they swim. Hagan (1943) states that a heavy loss of ducklings occurs on the duck ranches of Rhode Island where ducks are raised in large numbers on small areas of ground under poor hygienic conditions. Some evidence exists that eggs from infected birds may be a source of infection, since the organism of fowl cholera has been repeatedly found in them, yet doubt is cast on this theory from the fact that the disease is extremely rare in chickens under two months of age.

The possibility that insects may serve as vectors of fowl cholera has been considered, but such method of transmission is probably neither common nor important. However, Skidmore (1932) experimentally transmitted fowl cholera to turkeys by feeding flies that had previously fed on infected fowl cholera blood. He also showed that a drop of fowl cholera-infected blood placed on a glass slide, allowed to dry, and kept at room temperature proved infective after 8 days but not on the thirtieth day. Skidmore points out that chickens and turkeys are fly eaters, and that under natural conditions the ingestion of flies that have fed on fowl cholera-infected material might be the means of introducing fowl cholera into the flock. He recommends that when affected birds are destroyed, they should be killed in such a manner that blood will not be scattered on the premises. Van Es and Olney (1940) report that in their experimental yards, consisting of twelve lots more or less

continually occupied by an average of about 100 birds over a period of three years, during which time virulent outbreaks of fowl cholera were maintained in two of the lots, no appearance of the disease among the remainder of the poultry population ever occurred. The more virulent outbreaks in the cholera lots occurred at the height of the fly season, and though these lots were separated from adjoining ones only by an ordinary poultry netting fence, the disease never spread. In comparison with the transmission hazards of infection-carrying fowls, the part played by flies appears insignificant.

It is often difficult or impossible to determine how the disease is introduced into a flock (Pritchett and Hughes, 1932). Frequently it follows the addition of newly purchased stock to the breeding flock—or even the purchase of fowls for the table. The latter are particularly dangerous if their offal is fed to chickens with other garbage.

Such infection-carrier birds are of normal appearance and health, therefore unrecognizable by ordinary observation. Pritchett *et al.* (1930, 1932) in a study of several farm flocks in New Jersey, have shown the possibility of carrier birds spreading the disease. In one of these the healthy pullets saved for breeders were proven to be the reservoir of infection. These had become carriers by having passed through an outbreak of fowl cholera the previous year. Explosive outbreaks were the result of contact of healthy birds with these carriers. The infectious agent was isolated from the respiratory tract, and produced not only localized upper respiratory disease but typical cholera as well.

**Natural infection and pathogenicity.** It formerly has been held that natural infection with fowl cholera most commonly occurs through the intestinal tract. Recent studies cast considerable doubt on this belief. Hughes and Pritchett (1930) found it impossible to incite the disease by introduction of *P. avicida* into the alimentary canal, whereas it was readily accomplished by administration of the same strain into the upper respiratory passages. Hertel (1904) discovered that a duck which was fed the organs of fowls that had died from cholera had remained healthy, and that even after 58 days virulent *P. avicida* were present in its lungs. Müller (1910) could prove, with domestic fowls that had withstood a food infection, that the cholera bacilli remained at least four months in the various organs and could then be excreted in an infectious form. Hughes and Pritchett (1930) recovered *P. avicida* from a number of cases of spontaneous roup, rhinitis, and wattle disease. Hendrickson and Hilbert (1932) showed that *P. avicida* may be present in the blood stream of naturally infected birds for a period of at least 49 days prior to death; the organism multiplies rapidly in the body immediately preceding and following death. Webster (1930) concludes that the spread and severity of the disease are governed by the resistance of the host and the dosage of the organisms. His studies on the epidemiology of fowl

cholera also indicate that severe epidemic forms of the infection are associated with a relatively virulent type of organism which survives with difficulty in the host, whereas the endemic disease is caused by strains of relatively low virulence and high vegetative capacity.

Pasteur (1880b) had reported that the fowl cholera organism produces toxins in culture media, and it formerly was believed that a negative chemotaxis occurs because of this, thus accounting for the rapid increase of the organisms in the animal body. The investigations of Weil (1905) and others, however, proved that it is very questionable whether an extracellular toxin formation does occur. Hadley (1918) failed to demonstrate exotoxins in one of his most virulent cultures, and it is now held that the Pasteurella group, as a whole, is nontoxin-producing.

**Symptoms and course of the disease.** Van Es and Olney (1941) state that in a flock infected through natural exposure, 4 to 9 days usually elapse before appearance of symptoms, though explosive outbreaks have occurred in their experimental flocks within 48 hours after the introduction of infected birds. In outbreaks of the peracute form premonitory symptoms may be entirely lacking, and fowls will be found dead on the nest or beneath the perches. The rapid devastation of a flock in such outbreaks is indication of a virulence of causative organisms not surpassed or perhaps equaled by those of any other disease. Particularly is this true in the beginning of an outbreak, at which time it is not unusual to observe normal, healthy birds which within a few hours are dead. In a highly productive flock, apparently healthy in the evening, several dead birds are often found the next morning. Such an unanticipated outbreak of disease, particularly on premises where waterfowls are also affected, is good presumptive evidence of fowl cholera. So rapidly does the disease kill that sick birds may never be seen. Later in the progress of the disease the virulence of the causative organism decreases, and sick birds may be numerous, lingering for a few days, or assuming a chronic form. Such birds become stupid, refuse to eat or drink, and rapidly emaciate. They seldom recover, and those that do may become carriers of infection. Many become lame because of localization of the organism in the joints of legs or wings. From these inflamed joints *P. avicida* may be isolated. The wattles, too, may be the seat of infection, becoming edematous, hot, and painful. Difficult breathing indicates the presence of mucus in the respiratory passages. Diarrhea may or may not occur. In peracute cases it is unusual, because the birds do not live long enough for it to develop. In cases in which it does occur, the evacuations are copious, watery, and of a gray, yellow, or green color. These colorations are not necessarily indicative of cholera, because they occur in other types of enteric diseases. Although not an invariable symptom, the head and its adjuncts frequently become cyanotic. The comb and wattles may become a dull purple color.

   While the above-mentioned symptoms are highly suggestive of cholera, they cannot be considered wholly sufficient to warrant a diagnosis. Confirmatory evidence is possible only through bacteriological examination. Microscopic examination of the blood revealing typical bipolar staining organisms is the final evidence.

   **Lesions.** Post-mortem changes in fowls dying with cholera are generally neither characteristic nor constant. In peracute cases they may be altogether absent; in subacute, there are generally petechial hemorrhages in the mucous



Fig. 11.2. Hemorrhages on heart, gizzard and proventriculus in fowl cholera. (Biester, Iowa State College.)

membranes of the lungs and intestines. The serous surfaces may also be petechiated, and particularly the fat of the abdomen (Fig. 11.2). The section of the intestine showing the greatest inflammation and congestion is the duodenum. In the peritoneal cavity there is often found a cheesy exudate, resembling boiled egg yolk. A similar exudate may occur in the swollen joints of localized infection (Beach, 1922).

   The liver reveals a parenchymatous hepatitis, is light-colored but rarely green, is of firm consistency, often with light-colored, firm areas and streaks resembling cooked liver. Numerous pinhead necrotic foci may be distributed throughout the liver (Fig. 11.3). The spleen seldom shows changes, being

only occasionally congested and enlarged. The heart, especially of geese, has the surface covered with small, dark red blotches, and the pericardial sac may contain a quantity of fluid in which fibrinous flakes float.

In the localized form various parts show changes such as:

1. Catarrhal or rouplike form, with a tenacious exudate about the nostrils and beak, differentiated from roup in that some septicemic cases may occur, with a few cases dying more acutely than in roup.



FIG. 11.3. Foci of necrosis of liver in fowl cholera. Pasteurella found in lesions. (Biester, Iowa State College.)

2. Otitis media or wry neck form, in which a cheesy, yellow exudate occurs in the internal ear or in the bones at the base of the brain.

3. Edema of the wattles. This form is most common in males with large wattles. There is rapid swelling from no apparent cause, and the parts are red and hot.

4. Arthritic form, in which a yellow, cheesy exudate is deposited in the tendon sheaths, with swellings hot and painful.

5. Ovarian or peritonitis form. This is more acute than the arthritic form. A firm, cheesy, yellow exudate like cooked egg yolk surrounds the

ovary and may fill the peritoneal cavity. This is to be differentiated from ruptured ova due to traumatism.

Thorp *et al.* (1931) observed abscesses in the oviduct, suppurative arthritis of the coxofemoral articulation, and osteomyelitis of the proximal end of the femur in a group of spontaneously infected birds. Bacteriologic cultures from the heart blood and liver of this group of birds remained sterile. From three kidneys and three spleens, *P. avicida* was recovered, and also from the marrow of the femur and tibia of each of the ten birds studied.

**Diagnosis.** As previously suggested, a tentative diagnosis may be made from observation of the symptoms and lesions found, bearing in mind that none of these is sufficiently characteristic to warrant other than a presumptive diagnosis. One may be more definite in his conclusions if several birds of a flock are available for examination, since in the different specimens the various changes may be found which may be lacking in a single one. Only by microscopic and cultural methods can an accurate diagnosis be established. The diseases from which cholera must be differentiated are roup, fowl typhoid, and fowl pest.

In the first, the demonstration of the typical, bipolar organism of fowl cholera in the exudate and the more rapid, fatal termination of the disease are significant. In cases of acute disease in which the spleen is normal, fowl cholera may be assumed to be distinguished from fowl typhoid, which is characterized by an enlarged spleen. McNutt (1931) has stated that fowl typhoid is the only acute disease of fowls over two months of age that causes enlargement of the spleen.

The symptoms and rapid course of fowl pest and fowl cholera are so alike that differentiation is impractical. Microscopic and cultural examination and animal inoculation must be carried out. The rabbit, which is highly susceptible to infection with *P. avicida,* is quite refractory to the filtrable virus of fowl pest and is the most satisfactory laboratory animal to use for differential diagnosis of these two diseases.

**Immunity.** There is a great deal of contradictory evidence regarding production of artificial immunity. Pasteur, using a virulent culture of the fowl cholera virus attenuated by prolonged growth on artificial media, succeeded by vaccination in protecting fowls against subsequent exposure to the infection. His method, however, did not prove practical in the field, because a uniform attenuation could not be secured, and heavy losses sometimes occurred in vaccinated flocks. Hadley (1914) was encouraged by his experience in immunizing rabbits against the fowl cholera organism by use of a strain avirulent for fowls. His later attempts to induce immunity in fowls by the use of this living organism as a vaccine proved unsuccessful. Lignières and Lignières (1902) proposed a vaccine containing virulent organisms attenuated by heating to 43° C. for 5 days. This method, however,

was never adopted in practice. Mack and Records (1916) employed a vaccine of agar-grown organisms, killed with phenol 0.5 to 0.9 per cent, and reported results favorable in some flocks but unfavorable in others.

Van Es and Martin (1920) tested the efficacy of fowl cholera stock vaccines and bacterins, using 100 chickens treated with one to three injections. These were tested for immunity one to two weeks following the last injection, as were also 100 untreated chicks. The results were exactly the same in the two lots, 99 chickens dying in each.

Weil (1905) made laboratory tests of fowl cholera aggressin as a means of protecting susceptible fowls. While encouraging, as reported by him, no practical results followed.

Murray and McNutt, in a test of aggressin on a moderate-sized flock, one-half of which was injected and the other half held as checks, found that on subsequent exposure to feeding with the organs of fowls dead of cholera more birds succumbed in the treated than in the untreated lot.

Van Es and Olney (1936) had quite the same experience with aggressin used on 150 birds with 66 controls, the total mortality in the two lots being about equal. They conclude that there is no reason to believe that hemorrhagic septicemia aggressin can be depended upon to protect fowls actually exposed to *P. avicida* infection.

In Europe, passive immunization with fowl cholera antiserum prepared by hyperimmunization of the horse has found some favor. The immunity thus obtained is so short-lived (one to three weeks), however, that it is of doubtful practical use. In the United States, this method of attempted protection has never met with favor, due in part, perhaps, to the high cost of repeated injections found necessary.

**Prevention and control.** Since biological treatment for prevention and control of fowl cholera has proven so unreliable and unsatisfactory, other measures pertaining to the protection of the flock must be adopted. If the flock consists of several units, particular attention must be given to prevent the spread of the disease from the infected flock to the healthy. It is advisable to provide separate attendants for these. Proper flock management is conceded to be more effective and more economical than medicinal treatment of individual fowls. So many poultry diseases are not amenable to therapeutic treatment that it is a waste of labor and money to attempt it.

The soil of poultry yards is subject to gross contamination by the droppings of fowls, and in the case of such a disease as fowl cholera, these discharges are highly infectious. Since it is so difficult, if not impossible, to disinfect soil with chemicals to an appreciable depth, every effort is in order to keep the ground well cleaned. Badly infected soils will need be given time and opportunity for purification by the natural disinfecting agencies—air, sunshine, and the naturally present purifying soil bacteria. The latter, under

favorable conditions of moisture and heat, rapidly rid the soil of organic matter. Draining low places where water accumulates is in order. After an outbreak of disease the premises should be left unpopulated for a considerable period to permit biologic purification, and when repopulation is to be made, the safety of premises may be tested by placing a few fowls therein before risking a large number.

The natural feeding habits of fowls are such as to prevent the establishment of effective barriers to the spread of disease. The use of the most modern sanitary feeding and drinking utensils is urged. These should be so constructed and so placed as to avoid overflow of food and water on the soil or on the floor of the house to become contaminated with the infective droppings.

The food supplied an infected flock should be carefully controlled. If the ration is of high protein content this should be greatly reduced, since Beach has shown that such a ration is conducive to high morbidity and high mortality.

Literature records little, if any, well-substantiated evidence of the importance of flying birds as transmitters of disease, either mechanically or as infection carriers. It does appear reasonable, however, that such might be the case; therefore, such birds as pigeons, particularly, may well be prevented from associating with the flock, especially if fowl cholera is in the neighborhood.

The danger of infection being introduced by newly purchased stock or by return of exhibition fowls that have been at fairs or poultry shows has already been stressed, but merits reiteration.

Finally, the proper disposal of carcasses of fowls dead of cholera is of great importance. Fowls badly infected and without hope of recovery should be slaughtered without spilling blood, and their carcasses, together with those of the flock that have died, should be collected and carefully burned.

Following an outbreak it is advisable to clean and disinfect the house carefully. Walls, roosts, nests, and feeding and water containers may be scrubbed with very hot lye water, 1 pound lye to 30 gallons of water, followed by spraying with a 3 per cent creolin or lysol solution or 2 per cent formalin.

## REFERENCES

Beach, J. R.: 1922. Observation on the occurrence of fowl cholera in California. Poultry Sci. 1:186.
Gärtner, A.: 1898. Über das Absterben von Krankheitserregern im Mist und Kompost. Zeitschr. f Hyg. 28:1.
Hadley, P. B.: 1912. Studies on fowl cholera. R. I. Agr. Exper. Sta.. Bul. 150.
——: 1914. The reciprocal relations of virulent and avirulent cultures in active immunization. R. I. Agr. Exper. Sta., Bul. 159.
——: 1918. Studies on fowl cholera. V. Toxins of *B. avisepticus*. Jour. Bact. 3:277.
—— and Amison, E. E.: 1911. A biological study of eleven pathogenic microorganisms from cholera-like diseases. R. I. Agr. Exper. Sta., Bul. 146:52.
Hagan, W. A.: 1943. Infectious Diseases of Domestic Animals. Comstock Pub. Co., Ithaca, N. Y.

Hendrickson, J. M., and Hilbert, K. F.: 1932. The persistence of *Pasteurella avicida* in the blood and organs of fowls with spontaneous fowl cholera. Jour. Infect. Dis. 50:89.

Hertel, M.: 1904. Über Geflügelcholera und Hühnerpest. Arb. a.d. kaiserl. Gesundheitsamt. 20:453.

Hilbert, K. F., and Witter, W. F.: 1936. Report of the poultry disease laboratory at Farmingdale, Long Island. Ann. Rep. N. Y. St. Vet. Coll., 1934–35, p. 38.

Hinshaw, W. R.: 1924. Studies on poultry disease at feeding stations. Kan. Agr. Exper. Sta., Spec. Rep.

Hoffman, H. A., and Stover, D. E.: 1942. Analysis of 30,000 autopsies on chickens. Calif. Dept. Agr., Bul. 31:7.

Hughes, T. P.: 1930. The epidemiology of fowl cholera. II. Biological properties of *P. avicida*. Jour. Exper. Med. 51:225.

——— and Pritchett, I. W.: 1930. The epidemiology of fowl cholera. III. Portal of entry of *P. avicida:* reaction of host. Jour. Exper. Med. 51:239.

Hutyra, F., Marek, J., and Manninger, R.: 1938. Pathology and Therapeutics of Diseases of Domestic Animals. Alexander Eger, Chicago. I:105.

Kaupp, B. F., and Dearstyne, R. S.: 1925. The differential diagnosis of fowl cholera and fowl typhoid. Jour. Am. Vet. Med. Assn. 67:249.

Kitt, Th.: 1888. Beiträge zur Kenntniss der Geflügelcholera und deren Schutzimpfung. Deutsch. Zeitschr. f. Tiermed. 13:1.

Lignières, M.: 1900. Contribution a l'étude et de la classification des septicémies hémorrhagiques. Bul. Soc. Cent. Méd. Vét., Paris. 54:329.

Lignières, J., and Lignières, M.: 1902. La vaccination contre les pasteurelloses. Compt. Rend. Acad. Sci. 134:1169.

Mack, W. B., and Records, E.: 1916. The use of bacterins in the control of fowl cholera. Nev. Agr. Exper. Sta., Bul. 85.

Manninger, R.: 1929. Geflügelcholera in Handb. der Pathogenen Mikroorganismen (Kolle und Wasserman) 6(1):529–62.

McNutt, S. H.: 1931. Diagnosis of poultry diseases. The Iowa Veterinarian, April, 2:23.

Merchant, I. A.: 1946. Veterinary Bacteriology. The Iowa State College Press, Ames.

Müller, J.: 1910. Über die Ausscheidung virulenter Hühnercholerabakterien bei durchseuchten Tieren. Monatschrft. f. Tierheilk. 21:385.

Pasteur, L.: 1880a. Sur les maladies virulents et en particulier sur la maladie appelée vulgairement choléra des poules. Compt. Rend. Acad. Sci. 90:239, 952, 1030.

———: 1880b. De l'atténuation du virus du choléra des poules. Compt. Rend. Acad. Sci. 91:673.

Pritchett, I. W., Beaudette, F. R., and Hughes, T. P.: 1930. The epidemiology of fowl cholera. IV. Field observations of the "spontaneous" disease. Jour. Exper. Med. 51:249.

——— and Hughes, T. P.: 1932. The epidemiology of fowl cholera. VI. The spread of epidemic and endemic strains of *Pasteurella avicida* in laboratory populations of normal fowl. Jour. Exper. Med. 55:71.

Rosenbusch, C., and Merchant, I. A.: 1939. A study of the hemorrhagic septicemia pasteurellae. Jour. Bact. 37:69.

Salmon, D. E.: 1880. Investigations of fowl cholera. Reports of U. S. Comm. of Agr., p. 101.

———: 1881–82. *Ibid.*, p. 272.

———: 1883. *Ibid.*, p. 44.

Skidmore, L. V.: 1932. The transmission of fowl cholera to turkeys by the common house fly (*Musca domestica* Linn.) with brief notes on the viability of fowl cholera microörganisms. Cornell Vet. 22:281.

Thorp, Jr., F., James, W. A., and Graham, R.: 1931. An unusual form of fowl cholera. No. Am. Vet. 12:37.

Van Es, L., and Martin, H. M.: 1920. Value of commercial vaccines and bacterins against fowl cholera. Neb. Agr. Exper. Sta., Res. Bul. 18.

——— and Olney, J. F.: 1936. Immunizing value of commercial hemorrhagic septicemia aggressins. Neb. Agr. Exper. Sta., Res. Bul. 87.

——— and Olney, J. F.: 1940. Inquiry into the influence of environment on the incidence of poultry diseases. Neb. Agr. Exper. Sta., Res. Bul. 118.

——— and Olney, J. F.: 1941. Poultry diseases and parasites. Neb. Agr. Exper. Sta., Bul. 332.

Webster, L. T.: 1930. The epidemiology of fowl cholera. Experimental studies. I. Introduction. Jour. Exper. Med. 51:219.

Weil, E.: 1905. Untersuchungen über Infektion und Immunität bei Hühnercholera. Arch. f. Hyg. 52:412.

CHAPTER TWELVE

# TUBERCULOSIS

By WILLIAM H. FELDMAN, *Division of Experimental Medicine,*
*Mayo Foundation, Rochester, Minnesota*

☆   ☆   ☆

Tuberculosis of poultry may be defined as a contagious disease caused by *Mycobacterium tuberculosis avium.* The disease is characterized by its insidious chronicity, its long continuation in a flock when once established and its tendency to induce in infected birds a state of unthriftiness, decrease or stoppage of egg production, and finally death.

Although it was suspected that the disease in chickens is related to the disease in mammals, tuberculosis of chickens was not established definitely as a separate entity until 1883, a year after Koch's (1882) epochal announcement that tuberculosis of man and of cattle is due to a living bacterial parasite—the tubercle bacillus. Whether or not the microorganism of tuberculosis of chickens is identical with the microorganism responsible for tuberculosis of mammals provided much controversy, with Koch maintaining for many years that tubercle bacilli were always the same regardless of the species in which they might occur. However, Rivolta, and later Maffucci (1890), showed by convincing experimental procedures that the microorganism of tuberculosis of chickens is definitely dissimilar to that of bovine tuberculosis, and in 1901 Koch finally abandoned his previous position and declared that tuberculosis of poultry is unlike tuberculosis of human beings and that the disease in man is dissimilar to that of cattle. Consequently, it was settled finally that three different species of tubercle bacilli are concerned with tuberculosis of mammals and fowl. Differences in the three species, which can be recognized readily at the present time, were vague and confused for nearly twenty years after Koch had demonstrated that a specific microorganism is the cause of tuberculosis.

Although tuberculosis of chickens had been recognized as a contagious disease even before Koch succeeded in demonstrating the tubercle bacillus, the disease has continued to spread throughout most of the civilized world. With the available information on the nature of the disease, its eradication in the United States is entirely feasible. To fail to suppress the malady perpetuates an entirely unnecessary economic burden on American agriculture.

The more important reasons why tuberculosis of poultry should be eliminated may be stated briefly as follows: (1) affected birds are unthrifty; (2) tuberculous chickens are undesirable for human food; (3) diseased birds produce fewer eggs; (4) tuberculous chickens frequently are the source of tuberculosis of sheep and especially swine; and (5) avian tubercle bacilli are capable of sensitizing cattle to mammalian tuberculin.

The indictment against the tuberculous chicken is serious, and nothing has been offered to refute the fact that a tuberculous bird is an undesirable member of the farm economy and as such should be eliminated. Unfortunately, in many sections of the United States the disease is probably on the increase and we have failed as yet to formulate a comprehensive program that can be expected to eradicate the disease or even bring it under satisfactory control. The noteworthy results obtained in the program for the eradication of bovine tuberculosis should stimulate the attack on the disease in chickens. Unless avian tuberculosis is banished or at least reduced to an insignificant minimum, tuberculosis will continue to exact a heavy toll from the producer of chickens and the consumer of pork.

## INCIDENCE AND GEOGRAPHIC DISTRIBUTION

Tuberculosis of chickens is world-wide in its distribution, although the disease occurs more frequently in the North Temperate Zone than elsewhere. Although the disease exists in practically all of the United States, there is a marked difference in its incidence in different parts of the country. The highest incidence of infection occurs in flocks of the north central states where probably more than 50 per cent of the chicken population of the United States is found (Fig. 12.1). The states in which the disease is most prevalent include North Dakota, South Dakota, Nebraska, Minnesota, Iowa, Wisconsin, Illinois, Michigan, Indiana, and Ohio. Although no data are available as to what percentage of individual chickens in the states just mentioned are tuberculous, there are available figures that indicate that from 50 to more than 60 per cent of the flocks are diseased. Data available indicate that the incidence of the disease in the southern states is quite low. The explanation for this is not entirely obvious, although there are several possible contributing factors, such as climate, flock management, and duration of the infection. The maintenance of large flocks in the North and the practice of frequently acquiring new stock are probably important factors in the spread and continuation of the disease.

The difficulty of tuberculin-testing all chickens in the United States, or of testing even a majority of the flocks, makes it impossible to obtain exact data on what the incidence of tuberculous infection of chickens really is. However, figures are available as a consequence of various surveys that indicate in a general way the extent of the infection. Information obtained

from such sources indicates that the incidence of tuberculosis in poultry throughout the entire United States is probably between 5 and 6 per cent.

These figures may be misleading unless it is remembered that the incidence of infection varies greatly in different sections of the country. In the heavily infected north central states, where the disease is most prevalent, a conservative estimate would place the percentage of flock infection between 50 and 65, whereas in the southern states the percentage of infection is negligible (Fig. 12.1). Infection in the United States varies greatly in individual flocks from less than 5 per cent to as much as 95 per cent.
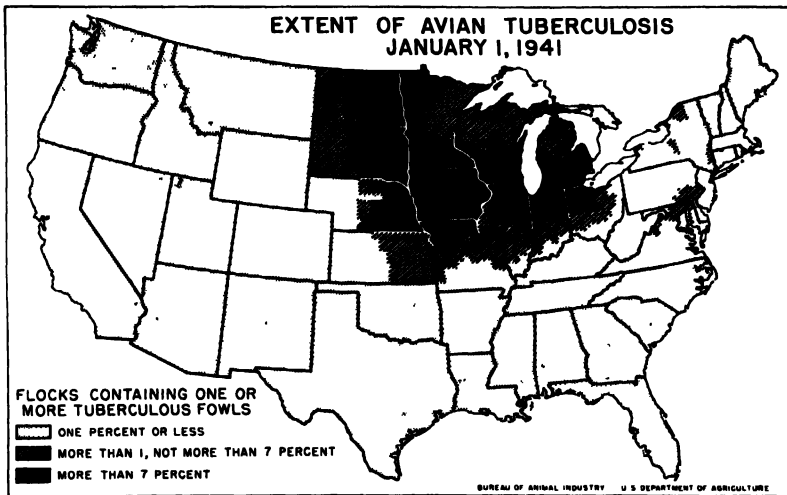


Fig. 12.1. Relative prevalence of tuberculosis of chickens in different areas of the United States. (Map kindly furnished by Dr. Elmer Lash, Bureau of Animal Industry, United States Department of Agriculture.)

As in the United States, the incidence of tuberculosis of chickens varies greatly in the different areas of Canada. From available data the incidence of infection in the different provinces varies from 1 per cent to 26 per cent. The disease occurs to only a limited extent in the Panama Canal Zone and has been observed but infrequently in Chile, Cuba, and Puerto Rico. The disease has not been reported from Colombia.

Considerable information concerning the incidence of avian tuberculosis in Europe is available. In Bulgaria the incidence of the disease is relatively low. The disease is rather prevalent in England and to probably a less extent in Scotland. Adequate information regarding the incidence of the disease in France is not at hand. However, the disease does exist in that country. Data from German sources prior to World War II indicate that avian tuberculosis was extremely prevalent in the German Reich and that the disease constituted a serious handicap to the poultry industry of that country.

Tuberculosis of chickens is practically unknown in Greece, infrequent in Switzerland, and apparently of rare occurrence in Italy. The disease occurs in Spain and seems to have assumed serious proportions in certain of the Baltic States. Avian tuberculosis is of frequent occurrence in certain districts of Norway. The disease occurs in Holland and is apparently on the increase in Czechoslovakia and Bessarabia.

Avian tuberculosis has been observed in a few instances in Dutch East Indies but is apparently rare or nonexistent in the Philippines. The disease had been noted in the Union of South Africa and New Zealand. In China there is a question whether the disease exists at all, although in Mukden there is evidence that indicates that the infection is fairly common.

**Age in relation to incidence.** Aside from the influence of climate and the factors of environment, the incidence of infection also depends upon the age of the chickens. This is well illustrated by the following data:[1] Twenty-eight farm poultry flocks in one county in Illinois were tuberculin-tested, and fourteen, or 50 per cent of the flocks, were found to contain tuberculous birds. In these flocks the incidence of tuberculosis among a total of 1,476 hens more than one year of age was 11.1 per cent. In 1,056 pullets (less than one year of age) in the same flocks, the incidence of infection as determined by the tuberculin tests was 0.19 per cent. Hays (1929) found among 40,073 chickens tested in Nebraska that the incidence of tuberculosis was 9.3 per cent. When the tuberculous birds were considered in relation to the different age groups, it was found that 77.6 per cent of the infected chickens were more than one year of age and 22.4 per cent were one year of age or less.

These data confirm the view that tuberculosis becomes more prevalent as the ages of the birds advance. There is evidence that indicates, however, that infection is less likely to succeed if exposure is delayed until the chickens reach the state of maturity, and that the disease when present in adult birds represents in most instances a process that began months or even years before, when the animal was young. Tuberculosis appears to be less prevalent in young fowl than in older birds, not because the younger birds are more resistant to infection than older birds, but because in the older birds the disease has had a greater opportunity to become established as a consequence of a longer period of exposure.

Although the lesions of tuberculosis in the young chickens are usually less severe than the lesions in the adult, extensive or generalized tuberculosis in young chickens has been observed occasionally. Such an animal obviously constitutes an important source of dissemination of virulent tubercle bacilli and must be considered a menace to other fowl and to susceptible mammals. That generalization of the disease does occur in young birds provides a

---

[1] Information supplied by H. R. Smith, Livestock Commissioner, National Livestock Exchange, Chicago.

formidable argument against the claim that avian tuberculosis can be eliminated by disposing of the older hens and maintaining a flock of young birds only. It is true that the concentration of potential infective materials will be diminished eventually if only young birds constitute the flock, but if the flock is maintained on contaminated premises new infections are likely, and the presence of even one pullet that has lesions along the intestinal tract will provide an adequate source of tubercle bacilli to insure the continuance of the disease.

### THE CAUSATIVE AGENT[2]

**Morphology.** The agent responsible for avian tuberculosis is a member of the genus Mycobacterium and is known correctly as *Mycobacterium avium.* The most characteristic feature of the organism is its acid-fastness.[3] The organism is capable of considerable pleomorphism, this feature being dependent on the character and chemical composition of the medium on which the bacteria are grown. While the organisms are bacillary in character, with perfectly straight forms usually present, clublike, curved, and crooked forms are also usually seen in most preparations. Branching infrequently occurs. Most of the bacteria have rounded ends. The bacteria vary in length from 1 to 3μ, and the average length has been determined to be 2.7μ. Spores are not produced, and the organism usually is considered to be nonmotile.

Reproduction of the avian tubercle bacillus is by simple fission. The existence of a filter-passing form of the avian tubercle bacillus has not been established convincingly. Spherical or conical granules occur in the endoplasm. These usually occur anywhere along the length of the bacterium. Whether or not the avian tubercle bacillus is capable of giving rise to forms comparable to "Much's" granules is uncertain. While the avian tubercle bacillus will grow and retain its virulence under a variety of atmospheric conditions, the organism is generally considered as aerobic.

The avian tubercle bacillus is not as exacting in its temperature requirements as are the human and the bovine forms of the organism. The avian form of the bacillus will grow at temperatures ranging from 25° to 45° C. although the most favorable temperature is between 39° and 40° C.

**Cultural distinctions.** The avian tubercle bacillus is not a difficult organ-

---

[2] Methods for the isolation and culture of tubercle bacilli are given in detail in a monograph on avian tuberculosis infections (Feldman, 1938).

[3] The Ziehl-Neelsen method of staining tubercle bacilli:

1. Cover a fixed film or smear preparation with Ziehl-Neelsen's carbolfuchsin (prepared by adding 10 cc. of a saturated alcoholic solution of basic fuchsin to 90 cc. of a 5 per cent aqueous solution of phenol). Steam gently but continuously for 2 minutes.

2. Wash in water and remove the uncombined stain by applying an excess of acid alcohol (prepared by adding 2 cc. of hydrochloric acid to 98 cc. of 80 per cent alcohol). Decolorization should be complete in 10 to 20 seconds.

3. Wash in water and counterstain with Löffler's methylene blue for 3 to 5 seconds.

4. Wash in water and dry. Examine with the oil immersion lens.

Tubercle bacilli and other acid-fast bacteria stain a bright red; other microorganisms stain blue.

ism to cultivate artificially. Stock strains will grow on most solid mediums although for original isolation of the organisms from naturally infected material one of the more specialized mediums is desirable. Glycerinated or nonglycerinated mediums are satisfactory, but the resultant colonies are larger if the medium contains glycerin than if it does not. After a culture has been obtained in solid mediums, subcultures usually will succeed in liquid mediums.

On mediums containing whole egg or egg yolk seeded with material prepared from tuberculous tissue and incubated at 37.5° to 40° C., the bacteria usually will become evident in 10 days to three weeks as small, slightly raised, discrete, grayish-white colonies. If the inoculation is rich in bacteria, the resultant colonies will be numerous and may tend to coalesce into granulated masses, but there is slight, if any, tendency for the individual colonies to spread. The colonies are hemispherical and do not penetrate into the substance of the medium. If the medium contains glycerin, the colonies gradually change from grayish-white to light ocher. The color becomes darker as the age of the culture increases.

Subcultures on solid mediums show evidence of growth within a few days and reach a maximal development in three to four weeks. Subcultures on solid mediums usually appear moist and unctuous, the surface eventually becoming roughened. The growth is translucent and has a creamy or sticky consistency; it is readily removable from the underlying medium. If the medium contains glycerin, the color finally becomes ocher.

In liquid mediums such as glycerinated broth, growth occurs at the bottom as well as at the surface. The surface growth is represented by a delicate filmy pellicle which, as growth continues, becomes thickened, slightly wrinkled, and granular. A "ring" of growth extending upward from the surface often occurs on the inner wall of the flask. Even after prolonged incubation, if the growth is not contaminated, the medium remains clear. The growth finally becomes golden yellow and gives off a characteristic odor.

While most strains of *Mycobacterium avium* are smooth and moist when first isolated, cultures of avian tubercle bacilli have been noted that were rough, dry, and crumbly. The occurrence of such strains, while not frequent, is sufficient reason for caution in designating the type of any tubercle bacillus without tests for pathogenicity.

Culturally, the avian tubercle bacillus is capable of dissociation. By this phenomenon marked variations in the physical character of the colonies are evident. The following variants have been noted: "S" or smooth, which produces a raised, smooth, moist growth; "FS" or flat and smooth, in which the growth is slightly wrinkled and moist and in which the colonies are larger than those of the "S" variant; "R" or rough, in which the colonies are large, dry, and wrinkled; and "CH" or chromogenic. The last-named is

physically not unlike the "S" variant except that the colonies become ocher in color.

In addition to the physical differences between the different variants, there also exist antigenic and pathogenic differences that are significant to a thorough understanding of the variation that occurs after natural infections with the avian tubercle bacillus. Experimentally, it has been noted that the "S" variant is far more pathogenic than the markedly dissimilar "R" variant, whereas the "CH" variant is essentially avirulent.[4]

**Stability of types.** Although nearly all strains of avian tubercle bacilli possess the essential cultural and pathogenic characteristics necessary to distinguish this organism from the other species of mycobacteria, occasionally strains have been encountered that appear to be atypical. The most likely explanation for the occurrence of such strains is that they represent variants that are themselves unstable. Some contend that the respective types of tubercle bacilli are not necessarily stable and that transmutation of one type into another may occur as a consequence of environmental adaptation. This is a controversial question, and while it remains unsettled the balance of evidence is strongly on the side of stability.

**Antigenic properties.** Although the human and the bovine forms of the tubercle bacillus are serologically indistinguishable by either the agglutination or the complement-fixation reaction, the antigenic structure of the avian tubercle bacillus is unlike that of the mammalian forms and may be distinguished from the latter by agglutinin absorption. However, avian tubercle bacilli do not constitute a homogeneous group. There exist at least three avian subtypes that can be distinguished serologically. Distinguishing precipitins have not been demonstrated.

### CHEMISTRY OF THE AVIAN TUBERCLE BACILLUS[5]

The avian tubercle bacillus contains carbohydrates, lipoids, and proteins, all of which, according to Sabin (1932), play a part in the cellular reactions of tuberculosis. Some of the carbohydrates are readily extractable; others occur in chemical combination, as, for example, the polysaccharides, manninositose, and trehalose, which are esterified with fatty acids. Renfrew (1929) has studied the carbohydrates which may be extracted with water from the defatted cells. The fraction, 1.4 per cent of the defatted cells, which may be precipitated with basic lead acetate, is smaller than the similar fraction from tubercle bacilli of the human type. Chargaff and Moore (1944) have isolated glycogen from avian tubercle bacilli by high speed centrifugation.

Renfrew reported the presence of about 10 per cent nitrogen in the

---

[4] A more complete discussion of dissociation and colony variation will be found on page 50 of monograph on avian tuberculosis infections (Feldman, 1938).
[5] This section was kindly prepared by Dr. Eunice V. Flock, Mayo Foundation.

defatted cells, of which 4.4 per cent is in water-soluble protein which possesses the characteristic biological activity of tuberculin, and 34 per cent is in protein, soluble in dilute alkali, with a much larger quantity remaining undissolved. Using another procedure, Menzel and Heidelberger (1938) isolated four fractions of proteins which gave a total of 14.2 per cent protein in the dried defatted cells. By serologic experiments, they demonstrated definite differences in specificity between the protein fractions of the avian and the corresponding protein fractions of the human and bovine types of tubercle bacilli.

The lipoids of the tubercle bacilli have been studied extensively by Anderson and co-workers (1942). With mild methods of extraction they (Anderson *et al.*, 1930a and b) found the lipoid content of dried avian tubercle bacilli to be 15.26 per cent; by more vigorous extraction, they (Anderson *et al.*, 1940) obtained an additional 10.8 per cent of firmly bound lipoids. The lipoids which are extracted by the mild treatment are phosphatides, acetone-soluble fat, and chloroform-soluble wax. The avian tubercle bacillus contains smaller quantities of phosphatides and acetone-soluble fat and a much larger quantity of the firmly bound lipoids than does the human type tubercle bacillus.

Like other tuberculophosphatides, those from avian tubercle bacilli contain low percentages of phosphorus and nitrogen (Anderson *et al.*, 1930a and b). The following fatty acids are found: palmitic, stearic, oleic, and a new group of saturated liquid fatty acids. The fatty acids are esterified with manninositose instead of glycerol. The saturated liquid fatty acids resemble tuberculostearic acid from the human strain in their biological activity. In the neutral fat, the fatty acids are esterified with trehalose.

The chloroform-soluble wax contains two optically active hydroxy acids of high molecular weight which have the property of acid fastness, (Anderson *et al.*, 1939) avian alpha and beta mycolic acid. They occur in combination with the carbohydrate trehalose. The unsaponifiable matter in this fraction is chiefly the higher alcohol *d*-eicosanol-2.

The firmly bound lipids (Reeves *et al.*, 1937) must be subjected first to acid hydrolysis before they can be extracted with the usual fat solvents. These lipids have a waxy appearance. The fatty acid, gamma mycolic acid, which occurs in this fraction, has a high molecular weight and is acid-fast (Anderson *et al.*, 1939). The neutral material in this fraction is *d*-eicosanol-2. In addition there is a specific polysaccharide which on hydrolysis yields mannose, *d*-arabinose, galactose, and traces of glucosamine and inosite.

### SUMMARY OF DISTINGUISHING FEATURES

The more essential features that distinguish avian tubercle bacilli from the human and bovine forms of the organism are summarized in Table 1.

Although in most instances differences in the physical properties are

sufficiently impressive to separate the avian from the mammalian forms of the tubercle bacillus, in critical work in which convincing proof of identification is desirable the animal inoculation method is the procedure of choice. In other words, the pathogenic behavior of a tubercle bacillus is more important that its physical characteristics in revealing whether or not the organism is the avian, human, or bovine form of *Mycobacterium tuberculosis*.

## PATHOGENICITY FOR OTHER FOWL

All species of birds are capable of being infected with avian tubercle bacilli. Some species are more susceptible than others, and generally speaking, those that are domesticated are affected more frequently than those

TABLE 1
SUMMARY OF ESSENTIAL DIFFERENCES BETWEEN AVIAN, HUMAN,
AND BOVINE FORMS OF THE TUBERCLE BACILLUS

|  | Avian | Human | Bovine |
|---|---|---|---|
| Growth in egg medium (without glycerin) | Grows readily. Culture moist and unctuous. Optimal temp. 40° C. | Grows well. Culture dry and roughened. Optimal temp. 37.5° C. | Grows slowly. Culture thin and without pigment. Optimal temp. 37.5° C. |
| Growth in liquid medium | Pellicle formation with crumbly granular growth at bottom | Pellicle formation with growth limited to surface | Pellicle with growth limited to surface |
| Miscibility with saline solution | Suspension easy. Organism uniformly distributed | Suspension difficult. Organisms form clumps | Suspension difficult. Organisms form clumps |
| Tuberculin sensitivity | More intense for homologous tuberculin | More intense for mammalian tuberculin | More intense for mammalian tuberculin |
| Pathogenicity * | Virulent for chickens and rabbits. Slightly pathogenic for guinea pigs | Nonpathogenic for chickens. Markedly virulent for guinea pigs but only slightly so for rabbits | Nonpathogenic for chickens. Markedly virulent for guinea pigs and rabbits |

* When testing tubercle bacilli for pathogenicity, chickens and rabbits should be inoculated intravenously; guinea pigs, subcutaneously.

living in a wild or free state. Among the domesticated fowl other than chickens in which tuberculosis may occur are turkeys, ducks, geese, swans, and peacocks. While tuberculosis occasionally develops in pigeons as a consequence of infection with avian tubercle bacilli, there is evidence that pigeons are more resistant to the infectious agent than are chickens. Parrots and canaries may be infected with avian tubercle bacilli, but it is the general impression that these species are less susceptible to avian tubercle bacilli than to the mammalian forms of the organism.

Among wildfowl, tuberculosis is infrequent. However, in wild birds that

frequent farm premises where tuberculosis is prevalent in chickens, the disease may be expected to develop. Pheasants seem to be markedly susceptible to infection by the avian tubercle bacillus, and the disease has also been observed in the sparrow, the crow, the barn owl, the cowbird, the blackbird, and the eastern sparrow hawk.

Tuberculosis is common among birds in zoological gardens. In the unnatural environment of captivity, the incidence of the disease frequently equals or even exceeds that for the domestic species of fowl. The infectious agent in practically all instances is the avian tubercle bacillus, although a few instances have been reported of birds being infected with heterologous strains of tubercle bacilli. Tuberculosis in the parrot is usually due to either the human or the bovine type of bacillus.

## PATHOGENICITY FOR MAMMALS[*]

The bacterium responsible for tuberculosis of fowl has a definite pathogenicity for some important species of domesticated mammals and at least a slight pathogenicity for others. This fact should be recognized more widely if the problem of eliminating tuberculosis infections is to be attacked intelligently and eventually solved.

Under conditions of natural exposure it is very exceptional for aggressive, extensive tuberculosis to develop in mammals other than rabbits and swine as a consequence of avian tubercle bacilli. Infection may occur, but the disease remains benign and localized. However, the microorganisms may assume a parasitic existence and multiply in the tissues for a considerable period, and in some instances may induce a state of sensitivity to tuberculin even though recognizable alterations in the tissues cannot be found. Although spontaneous infection of mammals fails in most instances to produce a disease of comparable severity to that which develops in fowl infected with avian tubercle bacilli, it is possible to produce extensive changes in many species of mammals by introducing the infective agent artificially.

**Guinea pig.** Naturally acquired avian tuberculosis in guinea pigs due to contact infection seldom has been observed. Guinea pigs are relatively resistant to avian tubercle bacilli, and while the resistance is not absolute the resultant disease is generally mild or limited, tends to heal, and rarely assumes an aggressive or generalized form such as characterizes infection with the human or bovine types of tubercle bacilli.

**Rabbit.** The rabbit is capable of being readily infected with avian tubercle bacilli experimentally. Tuberculosis may also develop as a consequence of natural or contact infection with the organism. If rabbits are permitted to occupy the same enclosure with tuberculous chickens or are kept in close

---

[*] A more complete consideration of the pathogenicity of avian tubercle bacilli for mammals will be found in the monograph by Feldman (1938).

proximity to chicken houses or runways where large quantities of tubercle bacilli are likely to occur in the soil and air-borne dust, the possibility of the dissemination of the disease to some of the rabbits is considerable.

**Rat.** While the rat is quite resistant to tuberculosis infection, a mild type of the disease, in which the organisms are capable of multiplying and remaining in the tissues for a considerable time, may follow intraperitoneal infection with avian tubercle bacilli.

**Mouse.** Only a few cases of natural infection of mice with avian tubercle bacilli have been reported. These animals have a relatively high tolerance for avian tubercle bacilli. Even though infected experimentally with large doses of virulent bacilli, the animals frequently live for many months before a lethal effect ensues.

**Dog.** Although moderately susceptible to the human and the bovine types of tubercle bacilli, the dog is highly resistant to infection with avian tubercle bacilli.

**Cat.** No instance of naturally acquired tuberculosis in the cat due to avian tubercle bacilli has been reported.

**Monkey.** Naturally acquired tuberculosis in monkeys due to avian tubercle bacilli is extremely rare. However, acute septicemic tuberculosis develops after the intravenous injection of avian tubercle bacilli. Monkeys are very refractory to infection by other routes of exposure.

**Sheep.** Sheep are moderately susceptible to infection with avian tubercle bacilli, and several cases have been reported, in some of which the disease has been severe and extensive.

**Goats.** Naturally acquired tuberculosis in goats is very infrequent, and no instance of such infection due to the avian tubercle bacillus has been reported in America.

**Horses.** Although tuberculosis is not a common disease in the horse, a considerable number of cases are on record. The infective organism was in the vast majority of cases the bovine tubercle bacillus. In a few instances avian tubercle bacilli have been the etiologic agents, but in America no cases of avian tuberculosis in the horse have been reported.

**Other mammals.** Naturally acquired tuberculosis due to the avian types of the tubercle bacillus has been observed in wild deer and in marsupials.

**Cattle.** Concerning the ability of avian tubercle bacilli to infect cattle, the significant information may be summarized as follows: (1) Avian tubercle bacilli have at least a limited pathogenicity for cattle. (2) The morbid changes produced are inclined to remain localized. Whether or not avian tubercle bacilli are ever responsible for destructive, widespread, or generalized tuberculosis in cattle is not known. (3) The avian tubercle bacillus is apparently capable of parasitic existence in the tissues of cattle without necessarily giving rise to recognizable tissue changes. However, when

lesions occur they show the histologic changes ordinarily recognized as tuberculosis. (4) Following natural exposure to avian tubercle bacilli, cattle may become sensitized to avian tuberculin, and some of the animals may react to mammalian tuberculin also. (5) The interpretation of the tuberculin test in cattle is made more difficult and uncertain by the existence of tuberculosis in poultry.

**Swine.** Swine are infected readily with avian tubercle bacilli, and the incidence of naturally acquired avian tuberculosis in swine represents an impressive amount of the total number of swine slaughtered annually in that part of the United States where the disease is most prevalent in chickens.

The ease with which swine are infected with avian tubercle bacilli requires that this phase of the avian tuberculosis problem be emphasized.

TABLE 2

INCIDENCE OF TUBERCULOSIS OF SWINE IN FEDERALLY SUPERVISED ABATTOIRS IN THIRTEEN
DIFFERENT CITIES FOR THE YEAR ENDING JUNE 30, 1946 *

| City | Percentage of Infection |
|---|---|
| Chicago | 7.65 |
| Cincinnati | 5.94 |
| Cleveland | 10 71 |
| Denver | 7 20 |
| Detroit | 11.42 |
| Ft. Worth | 2.38 |
| Kansas City | 3.99 |
| Omaha | 6.40 |
| St. Louis | 6 43 |
| San Francisco | 7.89 |
| Sioux City | 8.11 |
| South St. Joseph | 7.43 |
| South St. Paul | 15.31 |

* Data kindly supplied by Dr. A. R. Miller, Chief, Meat Inspection Division, Bureau of Animal Industry, United States Department of Agriculture.

Although sources of bovine tubercle bacilli have been all but eliminated,[7] it is disturbing to note that in 1940 an average of one out of every eleven hogs slaughtered under federal inspection in the United States was retained for tuberculosis (Smith, 1940). The failure of the elimination of tuberculosis of cattle to result in a satisfactory reduction of the incidence of tuberculosis in swine can be explained only by the fact that at the present time the vast proportion of tuberculosis of swine is due to infection derived either directly or indirectly from tuberculous chickens.

As may be noted in Table 2 the incidence of tuberculosis in swine subjected to post-mortem examination by the Federal Meat Inspection Service during the year ending June 30, 1939, varied in different cities from slightly

[7] At the present time (1947) the proportion of cattle affected with bovine tuberculosis in the United States is considerably less than 0.5 per cent.

over 2 per cent to slightly more than 15 per cent. The figures in most instances reveal a significant correlation between the areas where the incidence of tuberculosis of fowl is highest and the disease in swine presumably originating from such areas.

The fallacy of expecting tuberculosis of swine to disappear with the elimination of the disease in cattle is obvious. The disease will remain an unnecessary economic burden on swine husbandry until it is eliminated from chickens and other farmyard poultry.

**Human beings.**[8] The literature contains a considerable number of instances in which it was claimed that avian tubercle bacilli were responsible for a tuberculous infection in human beings. Very few of the published reports of such cases contain unequivocal proof necessary to substantiate the contention that avian tubercle bacilli were the agents responsible for the condition described. As a matter of fact in only a relatively small number of instances has the evidence necessary to prove an avian type of infection in man been presented. The rarity of such cases can only indicate that human beings are extremely resistant to this form of the tubercle bacillus.

## SOURCES OF INFECTION

In most instances the presence of tuberculosis in a farm flock can be explained by the fact that the flock is maintained in an unhygienic environment where infected birds have been kept for many years previously. Of course the introduction of new adult stock from sources where tuberculosis exists is a hazardous procedure and may be the means of transmitting the disease to a previously healthy flock. When new flocks are to be established from sources other than hatcheries, it is exceedingly important that the new stock be from sources where it is definitely known that tuberculosis does not exist. Another possible source of infection is uncooked garbage that may contain the offal from tuberculous fowl and trimmings from tuberculous swine carcasses in which the infectious agent was the avian tubercle bacillus.

Chicks hatched from eggs laid by hens naturally infected with tuberculosis are believed by some to constitute a factor in the transmission of the disease. However, at the present time there is no convincing evidence that tuberculosis is likely to be introduced into a flock in this manner. In other words, baby chicks, regardless of their maternal source, if reared in an environment free from tubercle bacilli, are unlikely to become tuberculous.

## SYMPTOMS

In attempting to recognize tuberculosis in the living bird, it should be borne in mind that but few symptoms of the disease are necessarily charac-

---

[8] A detailed discussion of avian tuberculosis in human beings has been published previously (Feldman, 1938, 1947).

teristic. From a practical point of view, probably the most expedient way of diagnosing the disease with certainty is by post-mortem examination. The lesions are fairly characteristic and not likely to be confused with other pathologic conditions. However, tuberculous fowl do manifest certain symptoms that, when considered with other factors, constitute presumptive evidence of the disease. It is seldom that any one infected bird will present all the symptoms that have been considered indicative of infection. However, if the disease be at all prevalent in a flock, several or most of the symptoms may be evident in different birds.

Since tuberculosis usually has a protracted course, the disease is more likely to be detected during life by careful and repeated examination than by a single cursory inspection. The incubation period is usually long, and since objective signs are absent in the early stages of the disease, it is usually not possible to diagnose the malady at that time unless resort be had to the tuberculin test.

Ordinarily, if the disease has progressed sufficiently to affect the physical condition of the bird, the animal will be less lively than its mates. If the disease be in an advanced stage the affected fowl fatigues easily and appears depressed or languid. Although the appetite usually remains good, there commonly occurs a progressive and striking loss of weight which often amounts to emaciation. The thinness of the tuberculous bird is especially noticeable in the muscles of the breast (Fig. 12.2). The pectoral muscles are often in a state of complete atrophy, and as a consequence the keel or breast bone becomes strikingly prominent and may be deformed. In extreme instances most of the body fat eventually disappears, and the face of the affected bird appears smaller than it would normally.

As the disease progresses the feathers assume a dull and ruffled appearance. The comb, wattles, and ear lobes often become anemic and thinner than normal, and the uncovered epidermis has a peculiar dryness. Occasionally, however, the comb and wattles have a bluish discoloration. Icterus, indicative of hepatic changes, may be noted.

Unlike tuberculous infections in many of the mammals, and contrary to the opinion of some writers, the disease in chickens apparently does not induce a febrile state. Even though the disease be most severe, the temperature of the affected bird remains within the normal range. In many instances the bird, when forced to move, reveals a unilateral lameness and walks with a peculiar jerky, hopping gait. This alteration of the gait, which appears to be characteristic, is probably due to tuberculous involvement of the bone marrow of the leg. Infrequently a wing may droop as though paralyzed owing to a tuberculous involvement of the humeral scapulo-coraccoid articulation. The lesion in this situation may rupture and dis-

charge thin or caseous material.  Paralysis due to tuberculous arthritis some-times occurs, but this is not a frequent symptom of the disease.

If the affected chicken be greatly emaciated, one may detect nodular masses along the intestine by palpation of the abdomen.  The great hyper-trophy of the liver of many tuberculous birds, however, may make this pro-cedure difficult or impossible.  In about 10 per cent of tuberculous chickens, one may recognize the disease by palpation of the involved thymus glands.  The crop should be empty if this procedure is to yield satisfactory results.



Fig. 12.2.  Carcasses of three tuberculous chickens from a flock in which the disease was rampant.  Note the extreme atrophy of the muscles of the breast.

Most tuberculous chickens have lesions along the intestinal tract, and if these be ulcerative, as they usually are, severe diarrhea that is usually un-manageable results.  The enteric disturbance includes an extreme weakness, and the affected bird assumes a sitting position as a result of exhaustion.

The duration of life of the tuberculous chicken is variable.  Affected birds may die within a few months or may live for years, depending on the severity or extent of the disease.  Death may occur from sheer exhaustion, or the affected bird may die suddenly as a consequence of hemorrhage from rupture of the affected liver or spleen.

But few of the symptoms given are necessarily characteristic of tubercu-losis.  Convincing proof of a tuberculous infection can be obtained best by necropsy.  In fact the importance of necropsy for establishing a diagnosis of·

tuberculosis in chickens cannot be overemphasized. However, in districts where tuberculous poultry exists, the presence of the disease is suggested by (1) unthriftiness, (2) progressive loss of flesh in spite of good appetite, (3) the chronicity of the symptoms, and (4) the occurrence of the disease in swine not exposed to mammalian tubercle bacilli.

Since the presence of tuberculosis does not preclude the co-existence of other diseases, there are certain conditions that must be differentiated from tuberculosis. These include neoplasia (tumors), tapeworm infection, entero-hepatitis, and certain arthritic conditions such as may be associated with fowl cholera, fowl typhoid, paratyphoid, and gout. From the standpoint of the pathologic findings, two facts should be borne in mind in distinguishing tuberculosis from other conditions of chickens. These are (1) the character and distribution of the lesions in the abdomen associated in a large percentage of cases with lesions in the bone marrow and (2) the presence within the morbid tissues of numerous acid-fast bacilli. The latter are especially significant since they do not occur in any other spontaneous disease of chickens.

### THE TUBERCULIN TEST

The method of choice for determining the presence of tuberculosis in the living chicken is the intradermal tuberculin test, which was first applied successfully to chickens by Van Es and Schalk (1914). When it is administered properly and the results are interpreted with understanding, the tuberculin test provides a satisfactory procedure for determining whether or not tuberculosis is present in a given flock. The test is not infallible, but in the hands of one competent to administer it the procedure offers an extremely valuable aid in the diagnosis and control of avian tuberculosis.

**Technic of test.** The equipment necessary consists of a sterile tuberculin syringe of the Luer type of 1 cc. capacity and a goodly supply of sterile hypodermic needles one-half inch (1.3 cm.) in length and of 25 to 26 gauge. Absorbent cotton and a few fluid ounces of 70 per cent alcohol should also be available. The tuberculin to be used should be that prepared for intra-dermic use from avian tubercle bacilli.[9] The bird should be restrained so that the head is entirely immobile. The site of injection is the wattle. If soiled, the surface of the wattle should be cleaned with alcohol; otherwise, cleaning or attempting to disinfect the skin is unnecessary. The operator grasps the wattle between the thumb and the forefinger of one hand, and with the other manipulates the syringe containing the tuberculin. The needle of the syringe then is inserted carefully into the lateral aspect of the dermis, and 0.03 to 0.05 cc. of tuberculin is forced into the tissue. If the

---

[9] Tuberculin prepared from mammalian strains of tubercle bacilli may elicit positive reactions in tuberculous chickens, but the results are generally unsatisfactory. More infected birds will be revealed with the avian product, and the reactions to avian tuberculosis are usually more pronounced than those elicited with mammalian tuberculin.

procedure has been accomplished properly, a small bleb or a small diffuse blanched area will appear where the tuberculin was deposited. Although fairly satisfactory results may follow if the tuberculin is injected into the subcutaneous tissue, it is a better practice in all instances to place the tuberculin intradermally.

**The reaction.** After 48 hours the chickens are examined and the results recorded. Using for comparison the opposite uninjected wattle, positive reactions are usually easy to recognize, although much experience and a thorough understanding of all factors involved are essential if the results are to be evaluated properly. A positive reaction is indicated by the presence in the tissues of the injected wattle of a soft swelling (Fig. 12.3). Not all reactions are of equal magnitude; some are small, and some result in a pronounced swelling which increases the thickness of the wattle one to five times. The swelling is due largely to edema which occurs in the zone of the connective tissue which lies between the layers of the reflected dermis. To a lesser extent the swelling is due to the increased width of the corium, which is filled with closely packed mononuclear histiocytic cells, a few eosiophilic granulocytes, and a variable number of lymphoid cells and lymphocytes (Fig. 12.4). If the reactions be severe the cellular response occurs throughout the corium of the entire wattle. Necrosis



FIG. 12.3. Positive tuberculin reaction in the wattle of a chicken 48 hours after intracutaneous injection of avian tuberculin.

of the tissues overlying the site of reaction rarely, if ever, occurs. Hyperemia of the region of the reaction is not apparent, and the swollen wattle is usually grayish or pale yellow. After 48 hours the swellings gradually subside and usually disappear within 5 days after the tuberculin was injected.

Certain aspects of the test may occasion confusion to some, and in interpreting the results one should keep in mind certain factors. Fairly frequently a negative reaction will result in a bird that is definitely tuberculous, and conversely, a positive result is sometimes obtained in chickens in which signs of tuberculosis cannot be demonstrated. In the latter instance, failure to find lesions of tuberculosis does not imply necessarily that tubercle bacilli are not present in the tissues of the chicken. If the disease is in an early stage, lesions are likely to be too small to be noted grossly or too few to be found by the ordinary methods of examination. In a satisfactorily large number of instances a definitely positive tuberculin test in chickens indicates that the bird has been exposed to avian tubercle bacilli. If a sufficiently diligent search be made by methods that are proper and adequate, the infective bacteria can usually be demonstrated in positive reactors.

Tuberculin is a bacteria-free substance prepared from the metabolic products of tubercle bacilli, and as used for the diagnosis of tuberculosis in chickens may be considered entirely harmless to tuberculous as well as to normal birds. Frequently the question is raised whether or not tuberculin injected into nontuberculous fowl may be responsible for a positive reaction on repetition of the test in the same bird. If retests are done after an interval of one month, false positive reaction will not occur. In other words, in
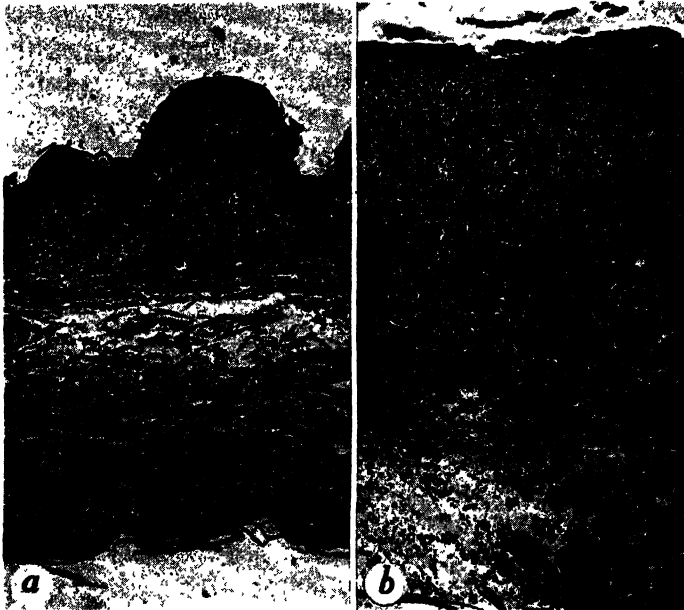


FIG. 12.4. a—cross section of an uninjected wattle showing the central connective tissue stroma and the epidermis of the opposite surfaces. ×56. b—markedly hyperplastic dermal tissue characteristic of a tuberculin reaction 48 hours after the injection of avian tuberculin. ×56.

chickens the usual diagnostic dose of tuberculin does not sensitize the nontuberculous animal to subsequent injections of the same product.

The tuberculin test has been utilized to a limited extent in diagnosing tuberculosis of turkeys. However, for the most part the results have been less satisfactory than for chickens. Certain difficulties are encountered also in tuberculin testing of pigeons and ducks. Generally speaking, the test is of limited value in diagnosing tuberculosis in these animals.

**Rapid agglutination test.** A serological procedure of possible diagnostic usefulness in tuberculosis of chickens has been suggested by the report of Moses, Feldman, and Mann (1943). By the use of concentrated suspensions of selected strains of avian tubercle bacilli, an antigen was prepared which was suitable for the detection of specific mycobacterial seroagglutinins. In

conducting the test, the procedure followed is essentially that of the rapid or plate agglutination tests.

Limited observations indicate that the agglutination test described has a diagnostic reliability in chickens comparable to the tuberculin test. The procedure should be subjected to more extensive trials since it offers certain distinct advantages. The animals need to be handled only once, and in addition, samples of serum submitted for agglutination tests for pullorum disease may also be examined for the presence of specific mycobacterium agglutinins.

### PATHOLOGIC ANATOMY

If a proper understanding of the disease problem as it affects the chicken flock is to be obtained, it is essential that a careful necropsy be made of all birds that die. Such an examination, if conducted by one who has knowledge of disease, will supply information that can be secured in no other way and will reveal the cause of death in a large percentage of instances. This is especially true in tuberculosis, in which the signs of the disease are fairly characteristic.

The gross morbid changes associated with tuberculosis of chickens that have died of the disease are usually strikingly evident. If the bird has died suddenly, one frequently find the abdomen filled with blood. The liver or spleen or both of these organs are greatly enlarged, and the source of the blood can be traced to rupture of one of these organs. Since birds suffering from leukosis may die as a consequence of rupture of the spleen or liver, additional evidence of tuberculosis must be sought in instances in which death was due to a fatal abdominal hemorrhage.

The pathologic changes in avian tuberculosis are those of infectious granuloma and, while the lesions have a general similarity to those of tuberculosis in mammals, there are certain characteristic distinctions.

It should be kept in mind that the character of the reaction of the tissues to the tubercle bacillus is not determined entirely by the character of the organism, but also by certain indefinite factors which are inherent in the species harboring the infection.

**Anatomic distribution of the lesions.** Since in the majority of instances tuberculosis of chickens is initiated by way of the digestive tract, it is not surprising that organs other than the lungs should show the greatest incidence of involvement. Lesions of the disease are seen most frequently in the liver, spleen, intestines, and bone marrow. The tuberculous bacillemia, which probably occurs intermittently and perhaps early in most if not all instances of tuberculous infection of chickens, provides a favorable circumstance for a widespread or generalized distribution of lesions. None of the tissues, with the possible exception of those of the central nervous system, appears to be immune from possible infection. Some of the organs such as the heart,

ovary, testis, and skin are affected infrequently and cannot be considered organs of predilection. In one series of 100 necropsies I found the lungs to be affected either grossly or microscopically in 48 per cent.

**Gross anatomy of the lesions.** Grossly, avian tuberculosis is characterized



Fig. 12.5. Tuberculous lesions in the liver of a naturally infected chicken.

by the occurrence of irregular grayish-yellow or grayish-white nodules of varying sizes in the organs of predilection such as the liver, spleen, intestine, and bone marrow (Figs. 12.5, 12.6, and 12.7). Involvement of the liver and spleen results in hypertrophy which is often of marked proportions. The
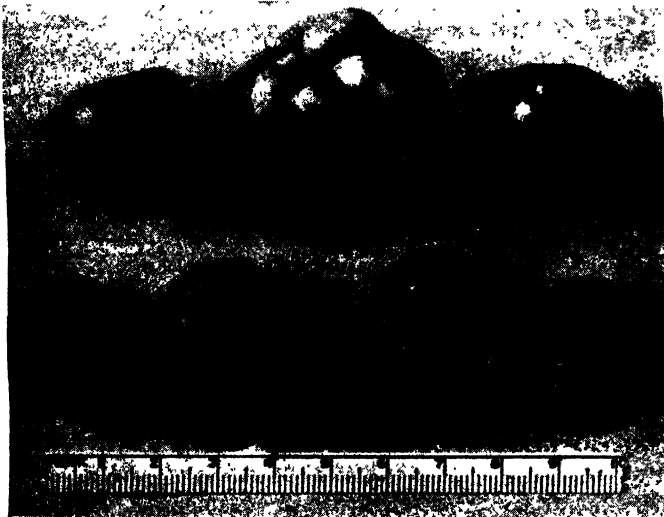


Fig. 12.6. Spleens from chickens naturally infected with tuberculosis. Note the variation in the number and size of the lesions.

tuberculous nodule, as observed grossly, varies in size from a structure that is just discernible to a huge mass that may measure several centimeters in diameter.[10] Nodules of large size frequently have an irregular knobby contour, and smaller granulations or nodules are often present over the surface. Lesions near the surface in such organs as the liver and spleen are enucleated easily from the adjacent tissues. The nodules are firm but are incised easily since mineral salts are not present. On cross section there may be observed a fibrous nodule containing a variable number of small yellowish foci or a single soft, yellowish central region which is frequently caseous. The latter is surrounded by a fibrous capsule the continuity of which often is interrupted by small circumscribed necrotic foci. The fibrous capsule varies in thickness and consistency depending upon the size and duration of the lesions. It is barely discernible or apparently absent in the smaller lesions and measures from 0.1 to 0.2 cm. in thickness in the larger nodules.

The number of lesions present is also variable, ranging from a few to innumerable. Large numbers of small lesions are particularly frequent in the liver and in the mesentery. It is fairly common to observe a few large nodular lesions in organs such as the liver and spleen associated with an enormous number of lesions of minute



FIG. 12.7. Large nodulated lesions of tuberculosis in the wall of the small intestine of a chicken.

to moderate size. The variation in the size of such lesions is a consequence of successive episodes of reinfection from previously established lesions, usually of the same organ. Involvement of the lung is usually less severe than of the liver or spleen (Fig. 12.8).

The continuous progressiveness of tuberculosis of chickens, once the disease is established, and the marked tendency of the disease to disseminate to several organs of the body indicate, as mentioned previously, that tuberculous bacillemia is a common manifestation of the disease. That the blood stream of tuberculous fowl does contain virulent tubercle bacilli at times has been demonstrated repeatedly. This tendency of the bacilli to invade, and circulate with, the blood stream provides the explanation for the frequent involvement of the bone marrow of tuberculous fowl (Figs. 12.9 and 12.10).

---

[10] Detailed descriptions of the gross and microscopic lesions of tuberculosis in the different organs of chickens will be found in monograph on avian tuberculosis infections (Feldman, 1938).

Infection of the bone marrow probably occurs very early in the course of the disease and is characterized by hypertrophy of the myeloid tissues, by disappearance of most of the bony spicules, and finally by the formation in the marrow of tuberculous nodules. The latter may be numerous and strikingly evident to the unaided eye, or the lesions may be few and of such size as to require the use of the microscope for their demonstration.

**Blood.** Reliable data on the effects of a natural tuberculous infection on the circulating blood of chickens are somewhat meager. Some workers have reported anemia associated with a reduction in the total number of erythrocytes. Other observations have indicated that there occur a marked increase in the number of large lymphocytes and a decrease in the small lymphocytes. The work of Olson and Feldman (1936) on a relatively small number of naturally infected chickens indicated that the erythrocyte and thrombocyte counts and the values for hemoglobin were within the limits of normal. Although in our material the disease was presumably of long duration, anemia was not observed. Leukocytosis was the most striking and consistent finding, the number of monocytes and heterophils being increased. The degree of leukocytosis was for the most part in direct ratio to the extent and severity of the disease.



Fig. 12.8. Large region of tuberculous involvement of one lung of a naturally infected chicken.

**Histopathology of the tubercle.** The anatomic unit of tuberculosis as the disease occurs spontaneously is conveniently designated a tubercle. The term "tubercle" as it refers to tuberculosis of fowl designates a structure which varies in character depending on its age and size. In its simplest form, which may be observed experimentally in 10 to 14 days after infection, there occurs a closely packed, microscopic collection of rather pale staining cells with vesiculated nuclei. These cells, which have been designated as epithelioid cells, contain tubercle bacilli and are derived from fixed tissue elements known as histiocytes (Fig. 12.11). The latter cells have a marked attraction for tubercle bacilli, which they phagocytose early in the reactive process.

The cellular mass or primary tubercle gradually expands as a consequence of the proliferative activity of histiocytes at the periphery, and within three to four weeks after the tubercle first becomes demonstrable, signs of retrogression can be detected in the epithelioid cells of the central zone. This retrogression is due in part to the avascularity of the structure and in part to the toxic substances of the tubercle bacilli. As the cellular mass becomes larger, the epithelioid cells have a tendency to fuse and form syncytia. The outlines of the individual cells become less distinct or disappear. Vacuoles



Fig. 12.9. Several femurs and one tibia from chickens naturally infected with tuberculosis showing lesions in the myeloid tissue and some well-marked osteoplastic changes due to the infection.

appear, and the staining reaction is more acidophilic. This is followed within a week or so by a necrobiotic change resembling coagulation necrosis The nuclei of the epithelioid cells become pyknotic and may disappear, while the cellular mass with the exception of the periphery becomes fused into a unit which stains deeply with eosin. The tubercle bacilli have multiplied and appear singly or in clumps throughout the necrotic tissue. This completes the first phase in the evolution of the tubercle.

The second phase of the development of the tubercle is concerned with the formation of giant cells. While the epithelioid cells in the central zone undergo necrobiotic changes, there persists an outer zone of epithelioid syncytia which appears as a mantle around the entire periphery. From these,

giant cells are developed. The giant cells thus formed may contain one or several nuclei. Infrequently, forms similar to Langhans' giant cells of mammalian tuberculosis may occur. The nuclei of the giant cells are situated distally to the central zone of necrosis, and the cells are arranged rather frequently in palisade formation. Large vacuoles often occur in the cytoplasm of the giant cells, and the nuclei stain intensely with the basic dyes. Immediately peripheral to the zone of giant cells there occurs a more or less diffuse collection of epithelioid cells and their progenitors, histiocytes, the latter being more numerous in the outer portion of the reacting zone (Fig. 12.12).
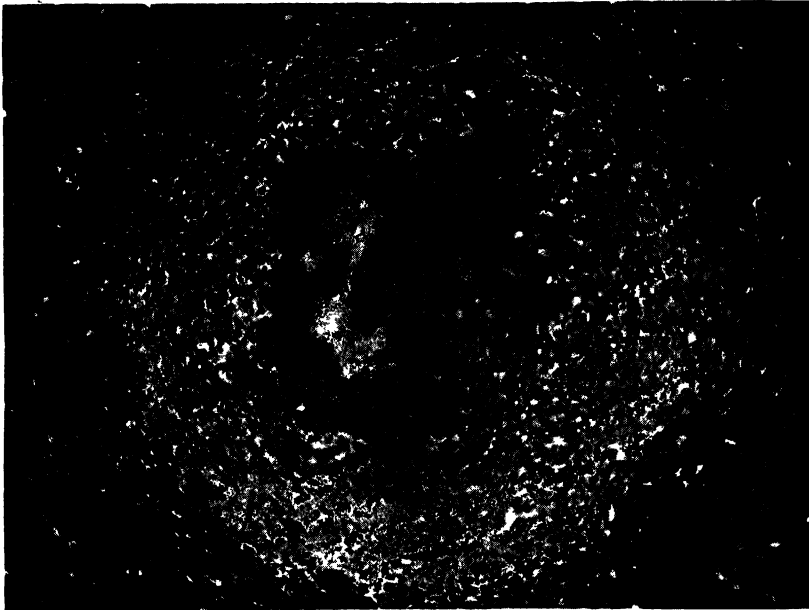


FIG. 12.10. Small tuberculous nodule in the bone marrow of a naturally infected chicken. The central necrotic region is surrounded by a zone of dense connective tissue. ×100.

Fibrocytes and minute blood vascular channels also occur near the outer portion of the peripheral reaction. While the tubercle bacilli are more numerous in the central or necrotic zone of the tubercle, they also occur in large numbers in the epithelioid zone, adjacent and distal to the giant cells.

The third and final phase in the formation of a tubercle is the development of a zone of encapsulation consisting of fibrous connective tissue, histiocytes, some lymphocytes and an occasional eosinophilic granulocyte. In limiting the progress of the disease the encapsulating structure is usually inadequate owing to the continuous development of new tubercles in the epithelioid zone immediately peripheral to the giant cells. As a consequence of these new or so-called daughter tubercles, a tubercle as recognized grossly

consists of the original or parent tubercle and several smaller or adjacent ones which considered together form a conglomerate tubercle (Fig. 12.13).

It is convenient morphologically to consider that the adult tubercle as it occurs in chickens consists of four parts or zones. The first is the necrotic or central zone, and the second the surrounding zone of giant cells. The third zone is that immediately peripheral to the giant cells and is composed of epithelioid cells and histiocytes. The fourth zone, which is not always apparent, is made up of histiocytes, small blood channels, and fibrous connective tissue elements.
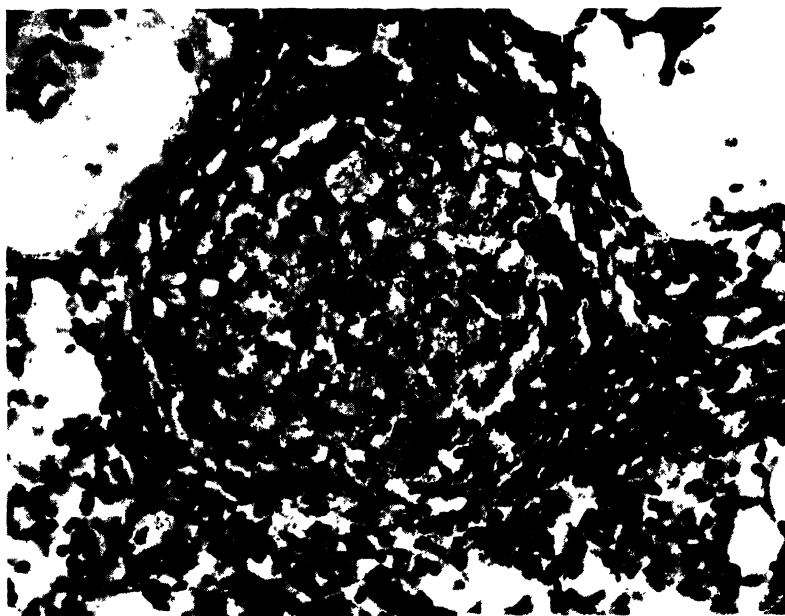


FIG. 12.11. Young epithelioid tubercle in the lung of a chicken. ×440.

The nature of the degenerative process which occurs in the central zone of the avian tubercle is somewhat unusual in that the integrity of the cells is maintained for a considerable period before disintegration becomes apparent. Caseation necrosis eventually occurs and may embrace all or a part of the central zone. Caseation probably is engendered by the influx of leukocytes, and there results a more or less structureless mass composed of tissue debris and nuclear fragments among which tubercle bacilli are numerous.

By appropriate stains, lipoid substances in variable amounts can be demonstrated in the lesions. The fat, which occurs in the form of small to moderately large globules, is most abundant in the more adult tubercles and of minimal amount in the prenecrotic epithelioid lesions.

In the first or epithelioid phase of the development of the tubercle, one

can demonstrate by appropriate staining methods the presence of delicate reticulum fibrils which intertwine promiscuously among the epithelioid cells. When degeneration and necrosis occur, the reticulum fibrils no longer can be seen.

Calcification of the tubercle occurs rarely if ever, the failure of mineral salts to accumulate being one of the unique characteristics of the tuberculous lesion as it occurs in fowl. Amyloid-like degeneration of portions of the surrounding parenchymal elements sometimes is observed in such organs as the liver, spleen, and kidney.

The number of tubercle bacilli present in the lesions is of much signifi-
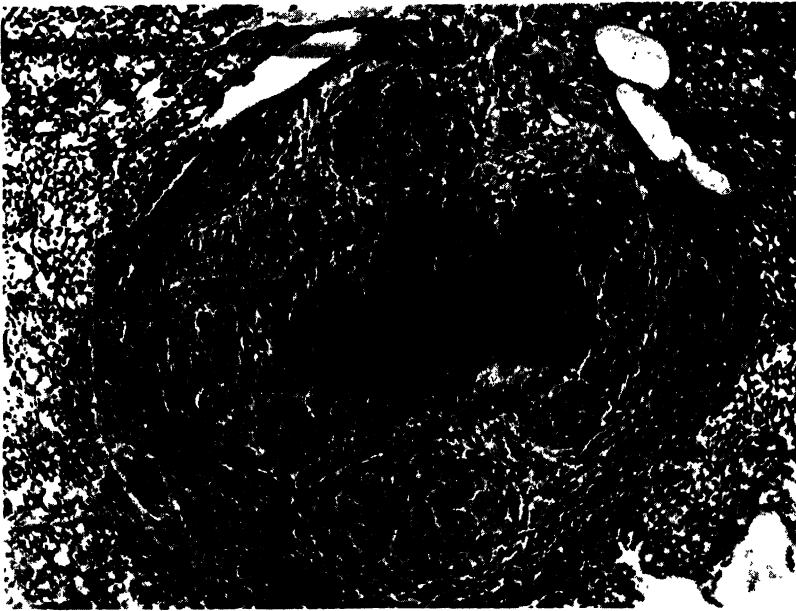


FIG. 12.12. Developing tubercle in the lung of a chicken showing activity of the third or tuberculogenic zone. ×100.

cance in the pathogenesis of avian tuberculosis. The propensity of the organism for growth and multiplication is hindered little if any within the tissues of chickens, and the results are prodigious numbers of bacilli in every lesion (Fig. 12.14). In this regard tuberculosis of chickens resembles two other mycobacterial diseases—leprosy and paratuberculosis. The organisms appear exceedingly numerous in smears from the morbid tissues, and cultures of tuberculous tissue from chickens usually yield innumerable colonies of tubercle bacilli. The presence of tubercle bacilli in such large numbers within the tissues of a tuberculous fowl constitutes an important factor in the transmission of the disease to healthy animals and makes

imperative the proper disposal of the carcasses of birds affected with tuberculosis.

Generally speaking, the morbid anatomy of tuberculosis of chickens is that of a serious, destructive, aggressive, granulomatous disease that seldom if ever heals spontaneously, and which in the great majority of instances eventually will result directly or indirectly in the death of the affected fowl.

### DISSEMINATION AND TRANSMISSION

Although several factors may contribute to the transmission and dissemination of avian tuberculosis to uninfected hosts, an infected environment is
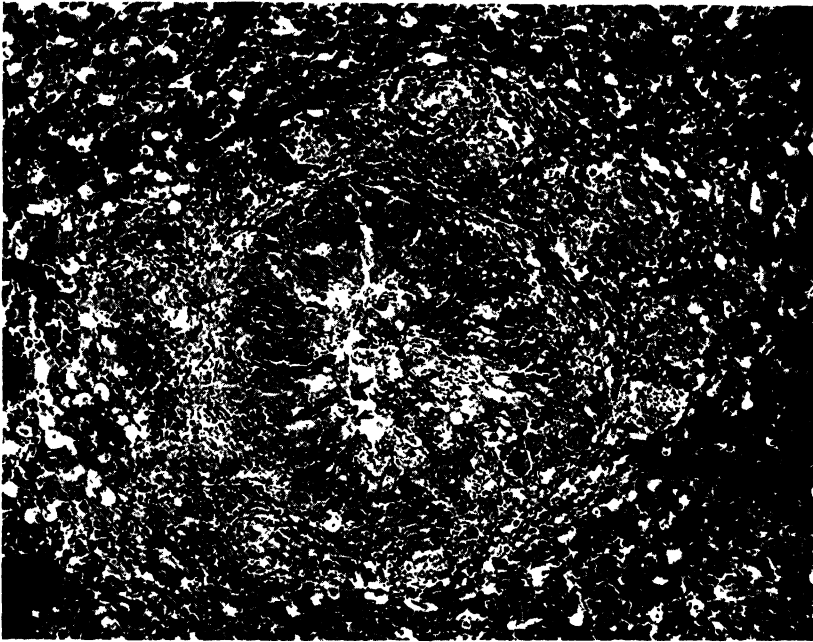


Fig. 12.13. Conglomerate tubercle in the lung of a chicken. Numerous secondary tubercles are present in the outer or peripheral part of the lesion. ×120.

the element of first importance in the perpetuation of the disease. Should a high percentage of infection occur as a consequence of an infected environ- ment, several related factors are of significance. These include (1) the age of the chicken, adult chickens being more resistant than younger ones; (2) the concentration of the infective material, premises occupied by many tubercu- lous fowl over a period of years being a more potent source of infection than premises that have been contaminated recently by relatively few tuberculous fowl; (3) repeated episodes of exposure over a considerable period; and (4) the complex and little understood question of individual susceptibility or resistance.

Of primary importance in the establishment of an infected environment are certain distinctive factors characteristic of the pathology of avian tuberculosis in the natural host. As mentioned before, the tremendous number of tubercle bacilli exuded from the frequently occurring ulcerated tuberculous lesions of the intestine establish the infected bowel as a constant source of virulent bacteria. These mix with the intestinal contents and eventually leave the body with the feces. Although other potential sources of infection exist, there is none that equals infective fecal material in the dissemination of
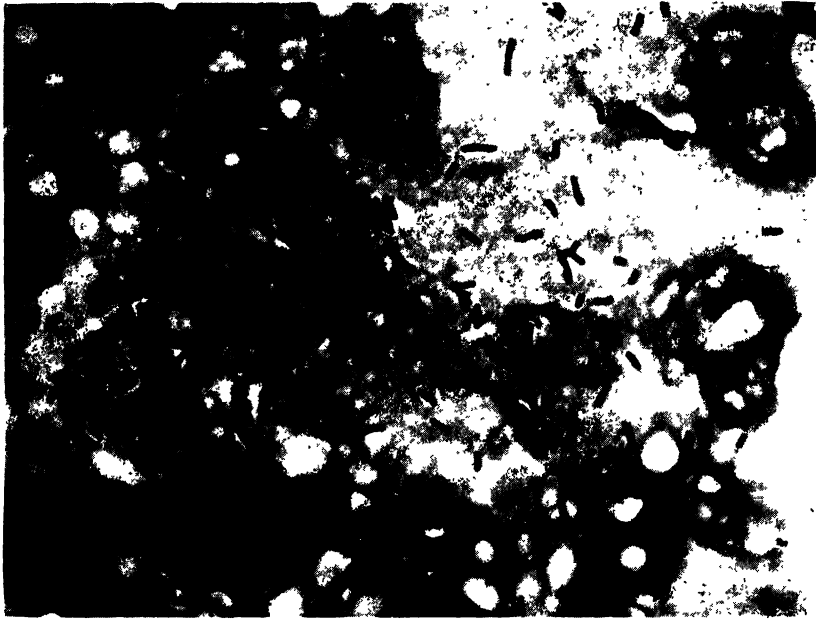


Fig. 12.14. Numerous tubercle bacilli in a smear preparation from a small lesion of the lung of a naturally infected chicken (stained by the method of Ziehl-Neelsen). ×1,600.

avian tuberculosis from affected to nontuberculous animals. Related to enteric ulcerations as sources of tubercle bacilli that may occur in the fecal discharges are lesions of the liver and of the mucosa of the gall bladder. From such lesions the organism fairly commonly finds its way into the intestine by way of the common duct.

Elimination of the bacteria from the respiratory tract is also a potential source of infection especially if lesions occur in the tracheal mucosa in addition to the lungs. The auto-ingestion of infective exudates by a bird that has tuberculosis of the respiratory tract also provides a potent source of re-infection with the possibility of the establishment of additional lesions in the other organs, especially the intestines.

**Vectors.** The possibility that living foreign-host carriers may transport avian tubercle bacilli from infected to noninfected premises constitutes an

interesting phase of the epidemiologic aspect of avian tuberculosis. Many have studied the problem, the report of Schalk, Roderick, Foust, and Harshfield (1935) being especially noteworthy. Although a resumé of the facts indicates quite definitely that vectors have a role in the dissemination of avian tubercle bacilli from infected to healthy flocks, it is hardly likely that vectors are responsible for any considerable amount of the tuberculous infection that exists in the average farm flock. They are perhaps more important as possible sources of new foci of infections in premises that were previously free of the disease. The infective environment, comprising as it does the bacilli-laden soil, litter, and filth, is the factor of greatest importance in the transmission of the disease to noninfected animals. The longer the premises have been occupied by infected birds and the more concentrated the poultry population, the more prevalent the infection is likely to be.

**Role of eggs.** The possibility that avian tuberculosis might be transmitted through the eggs from tuberculous hens has been a pertinent question for the past fifty years. Sibley (1890) observed the occurrence of tuberculosis in chickens that had been hatched from eggs laid by hens affected with the disease. In attempting to explain the origin and continuation of the disease Sibley stated that "the disease appears to be a clear case of heredity."

Generally speaking, the problem concerning the possible transmission of avian tuberculosis through infected eggs has been approached in two ways: (1) by inoculating eggs artificially with tubercle bacilli and noting whether or not tuberculosis eventually develops in the birds hatched from the infected eggs and (2) by observing whether or not tuberculosis develops in chickens hatched from eggs obtained from naturally infected hens.[11] Although it has been demonstrated many times that a portion of the eggs artificially inoculated with avian tubercle bacilli will hatch and that there is a good possibility that the chicks hatched from such eggs will be infected with tubercle bacilli and further that such infected chicks usually will die within a short time of extensive tuberculosis, from a practical point of view such observations are of questionable importance to the fundamental question: Are eggs from naturally infected chickens likely to produce chicks that are destined to be tuberculous? The information presented up to the present time is inadequate to support the belief that eggs from tuberculous chickens constitute a factor of importance in the dissemination of tuberculosis from infected to healthy flocks. Although the possibility that this might occur is admitted there is at the present time no convincing experimental evidence to justify the conclusion that chicks hatched from eggs laid by tuberculous hens will be infected with tubercle bacilli as a consequence of the infectious agent having been implanted in the developing embryo during the prenatal existence of the chick.

---

[11] Fitch, Lubbehusen, and Dikmans (1924), as a result of a comprehensive study, concluded that viable tubercle bacilli are present in less than 1 per cent of eggs from tuberculous chickens.

The most convincing evidence that infection from a tuberculous maternal parent is not likely to occur is that furnished by investigators such as Schalk, Roderick, Foust, and Harshfield (1935), and Fitch and Lubbehusen (1928) who reared many hundreds of chicks hatched from eggs of naturally infected hens without tuberculosis having been observed in a single instance.

**Other sources.** Another potent source of dissemination of avian tubercle bacilli is the bodies of tuberculous fowl that died of the disease or the offal from chickens that, although tuberculous, were dressed for food. It is obvious that any tissue likely to contain living avian tubercle bacilli should be disposed of in such a manner as to preclude its being eaten by chickens or swine. Infected tissues preferably should be burned or, if the food value of such flesh be of sufficient importance, it should be cooked thoroughly before being used as food for animals.

It is also conceivable that cannibalism might play a part in the transmission of tuberculosis from one chicken to another. Since bacillemia is of frequent occurrence in the natural course of tuberculosis of chickens it is reasonable to believe that the fierce and bloody assault on an infected bird by one addicted to cannibalism would provide a possibility that the aggressor would ingest tubercle bacilli with the blood of the infected victim. Whether or not such exposure would be sufficient to produce tuberculosis is problematic.

Avian tubercle bacilli may be transmitted from one situation to another by persons whose shoes may become soiled with fecal matter and other filth of the poultry yard. The equipment used in the care and maintenance of infected poultry flocks, such as crates and feed sacks, also might be responsible for the transfer of the infective bacteria from diseased to healthy flocks.

### CONTROL OF AVIAN TUBERCULOSIS

The eradication of avian tuberculosis or even its satisfactory control is not a simple matter. However, the widespread distribution of the disease, its high incidence in the more seriously infected areas, and the increasing importance of the poultry and the swine industries make it imperative that adequate measures be devised for its control and ultimate suppression.

As in most other infectious diseases, vaccination for the prevention of avian tuberculosis has been considered and tried. The products used include the so-called Friedmann vaccine,[12] an avian strain of BCG, and heat-killed avian tubercle bacilli. The result obtained from any of these products would not justify the claim that avian tuberculosis as the disease occurs naturally can be controlled successfully by vaccination. A measure of resist-

---

[12] Prepared from a so-called turtle strain of acid-fast bacilli (*Mycobacterium chelonei*).

ance can be conferred by the use of homologous strains, but much additional work will be necessary before vaccination can be accepted as worthy of serious consideration in the prevention of the disease.[13]

The tuberculin test if used judiciously is of considerable practical value in reducing the losses from tuberculosis. The subsequent removal from the flock of chickens that react eliminates many foci of infection. The test enables one to detect many infected fowl before the disease reaches a severe or chronic state, and if repeated tests are made, potential dissemination of the infective bacteria to the surrounding environment may be reduced appreciably. However, this method, if depended on alone for combating avian tuberculosis, has many shortcomings, the most important being that if the residual flock is permitted to occupy the same infective premises a rather constant source of infection remains. This provides opportunity for new infections to occur for an indefinite period, since in the soil, avian tubercle bacilli may remain viable and virulent for years. For this reason an environment once infected remains a potential source of infection indefinitely. Furthermore, neither the tuberculin test nor any other means can be depended on with absolute certainty to detect every living tuberculous fowl, and as long as one infected bird remains in a flock, dissemination of the disease to healthy fowl is possible. Consequently, means other than the tuberculin test must be resorted to if a more satisfactory control of avian tuberculosis is to be expected.

For the past several years it has been generally stated that avian tuberculosis can be controlled if all birds in the flock be disposed of after the first laying season. This practice has much to commend it, especially since it is economically sound from the point of view of egg production. Older birds usually produce fewer eggs than the younger ones, and furthermore, the mortality from nonbacterial diseases such as neoplasia is greater among the older hens than among pullets. Another factor in favor of the disposal of the older stock is that if tuberculosis is present it is usually more severe in the older birds which are as a consequence more likely to become depots of dissemination.

Desirable as it may be to dispose of the older birds, to maintain that such a practice will rid the flock of tuberculosis is at best an optimistic wish. Contrary to the belief of some, acute generalized tuberculosis occasionally occurs in pullets. The lesions in such a bird contain enough tubercle bacilli to infect a dozen flocks, and if the carcass be eaten by its mates the likelihood that several additional birds will become infected is evident. To reiterate, the threat of avian tuberculosis in potentially serious proportions remains just so long as a single infected bird is a member of the flock. The removal

[13] The question of vaccination against tuberculosis of chickens is reviewed in monograph on avian tuberculosis infections (Feldman, 1938).

of all birds after the first laying season is a practice not without merit, but that avian tuberculosis can be eliminated by this means remains to be demonstrated.

Since it has been adequately established that the continuation of tuberculosis in a flock is dependent on an infected environment, it would seem reasonable to believe that this fundamental fact should be utilized in any program devised for the control and elimination of the infection. Basically the question is one of hygiene. That every case of tuberculosis comes from another case is aphorismic. After all, the disease is due to a well-known and definitely established cause, the tubercle bacillus, and this fact must not be lost sight of or ignored when measures to eliminate the infection are considered. To permit young birds, even though free from the disease, to range at will over premises that are infected or to occupy quarters that were used previously to maintain tuberculous birds is to insure the continuation of the disease indefinitely.

Procedures for establishing and maintaining tuberculosis-free flocks should embrace the following: (1) Abandon the old equipment and establish other facilities on new soil that is known not to be contaminated with avian tubercle bacilli. Ordinarily it is impractical to render an infected environment satisfactorily safe by disinfection. (2) Provide proper fencing or other measures to prevent the unrestricted movement of the chickens, thus preventing exposure from previously infected premises. (3) Eliminate as soon as possible the old flock, burning the carcasses of birds that show lesions of tuberculosis. (4) Establish a new flock in the new environment from tuberculosis-free stock. (5) Eliminate from the swine herd all reactors to avian and to mammalian tuberculin. New breeding stock should likewise be tuberculosis-free. If the chickens in such a flock are prevented from having access to an infected environment and are protected against accidental exposure to tubercle bacilli, it is reasonable to believe that they will remain free from tuberculosis.

The measures just mentioned for the elimination of avian tuberculosis are not complicated and should be applicable to most American farms. The additional profits that will accrue from a tuberculosis-free flock maintained in a hygienic environment will in time compensate adequately for the initial expense and work necessary to establish the new flock and new facilities. Furthermore, the general health of the birds will be better, and diseases other than tuberculosis will be controlled more satisfactorily. The benefits will also be reflected in a decrease in tuberculosis in swine. The importance of avian tuberculosis in the infection of swine is such that if chickens were maintained entirely separate and apart from swine, the incidence of tuberculosis of swine would be reduced to a minimum.

## ACKNOWLEDGMENT

## REFERENCES

Anderson, R. J.: 1942. The chemistry of the lipids of the tubercle bacillus. Yale Jour. Biol. and Med. 15:311.

—— and Creighton, M. M.: 1939. The chemistry of the lipids of tubercle bacilli. LVII. The mycolic acids of the avian tubercle bacillus wax. Jour. Biol. Chem. 129:57.

——, Creighton, M. M., and Peck, R. L.: 1940. Chemistry of lipids of tubercle bacilli. LX. Concerning the firmly bound lipids of the avian tubercle bacillus. Jour. Biol. Chem. 133:675.

—— and Roberts, E. G.: 1930a. The chemistry of the lipoids of tubercle bacilli. X. The separation of lipoid fractions from avian tubercle bacilli. Jour. Biol. Chem. 85:509.

—— and Roberts, E. G.: 1930b. The chemistry of the lipoids of tubercle bacilli. XI. The phosphatide fraction of the avian tubercle bacilli. Jour. Biol. Chem. 85:519.

Chargaff, E., and Moore, D. H.: 1944. On bacterial glycogen: the isolation from avian tubercle bacilli of a polyglucosan of very high particle weight. Jour. Biol. Chem. 155:493.

Feldman, W. H.: 1938. Avian Tuberculosis Infections. The Williams and Wilkins Company, Baltimore, p. 483.

——: 1947. Animal tuberculosis and its relationship to the disease in man. Ann. N. Y. Acad. Sci. 48 (Art. 6) :469.

Fitch, C. P., and Lubbehusen, R. E.: 1928. Completed experiments to determine whether avian tuberculosis can be transmitted through the eggs of tuberculous fowls. Jour. Am. Vet. Med. Assn. 72:636.

——, Lubbehusen, R. E., and Dikmans, R. N.: 1924. Report of experimental work to determine whether avian tuberculosis is transmitted through the eggs of tuberculous fowls. Jour. Am. Vet. Med. Assn. 66:43.

Hays, C. H.: 1929. Avian tuberculosis in Nebraska. Jour. Am. Vet. Med. Assn. 75:549.

Koch, R.: 1882. Die Aetiologie der Tuberkulose. Berliner klin. Wochenschr. 19:221.

Maffucci, A.: 1890. Beitrag zur Aetiologie der Tuberkulose (Hühnertuberkulose) . Zentralbl. f. allg. Path. u. path. Anat. 1:409 (June 15) .

Menzel, A. E. O., and Heidelberger, M.: 1938. Cell protein fractions of bovine and avian tubercle bacillus strains and of the timothy-grass bacillus. Jour. Biol. Chem. 124:301.

Moses, H. E., Feldman, W. H., and Mann, F. C.: 1943. Mycobacterial rapid agglutination antigens and their diagnostic value in tuberculosis of fowl. Am. Jour. Vet. Res. 4:390.

Olson, Jr., C., and Feldman, W. H.: 1936. The cellular elements and hemoglobin in the blood of chickens with spontaneous tuberculosis. Jour. Am. Vet. Med. Assn. 89:26.

Reeves, R. E., and Anderson, R. J.: 1937. The chemistry of the lipides of tubercle bacilli. XLVII. The composition of the avian tubercle bacillus wax. Jour. Am. Chem. Soc. 59:858.

Renfrew, A. G.: 1929. A proximate analysis of a defatted residue of avian tubercle bacilli. Jour. Biol. Chem. 83:569.

Rivolta: Quoted by Maffucci, A.

Sabin, F. R.: 1932. Cellular reactions to fractions isolated from tubercle bacilli. Physiol. Rev. 12:141.

Schalk, A. F., Roderick, L. M., Foust, H. L., and Harshfield, G. S.: 1935. Avian tuberculosis: collected studies. N. D. Agr. Exper. Sta., Tech. Bul. 279.

Sibley, W. K.: 1890. Tuberculosis in birds. Jour. Comp. Med. and Vet. Arch. 11:317.

Smith, H. R.: 1940. Statistical tables showing progress in eradicating tuberculosis from livestock. Compiled from records of United States Bureau of Animal Industry. U.S.D.A.

Van Es, L., and Schalk, A. F.: 1914. Avian tuberculosis. N. D. Agr. Exper. Sta., Bul. 108.

CHAPTER THIRTEEN

# INFECTIOUS CORYZA

By J. R. BEACH, *Department of Veterinary Science,*
*University of California, Berkeley, California*

☆   ☆   ☆

Coryza, or colds, is probably the most commonly occurring and wide-spread of all diseases of poultry. In much of the early literature the disease was referred to as "roup," a term which has largely been discarded by poultry pathologists, because the term "roup" had no scientific or medical basis, and its use led to confusion since it was applied to the diphtheritic lesions of fowl pox involving the oral and ocular mucous membranes as well as to coryza. The frequent occurrence of the disease in company with other infections, particularly fowl pox, led early investigators to the erroneous conclusion that it was a part of the disease which it accompanied. For many years, therefore, a specific causative agent for coryza escaped detection. Conse-quently, environmental conditions such as dampness, sudden changes in temperature, crowding, and poor ventilation were regarded as necessarily predisposing or primary causative factors of the disease, and the micro-organisms present in the nasal chambers of affected chickens were presumed to play a secondary etiological role. During the period from 1932 to 1936, however, several investigators, widely scattered and working independently, succeeded in isolating from the nasal exudate an organism which, when in-jected into the nasal passages of healthy chickens, would reproduce the symp-toms of the natural disease. This type of respiratory infection, to which the term infectious coryza has been applied, is the one to be discussed in this chapter. Infectious coryza is known to affect many flocks, but it is by no means certain that all uncomplicated epidemic or endemic coryza of chickens is etiologically identical.

**Etiology.** During 1932–36, de Blieck (1932, 1934), Nelson (1933a, 1933b), Delaplane, Erwin, and Stuart (1934), Delaplane (1936), Eliot and Lewis (1934), Schalm and Beach (1934, 1936a, 1936b), and Beach and Schalm (1936) reported the isolation from field cases of coryza in Holland, New Jersey, Connecticut, Maryland, and California, respectively, of a pleo-morphic hemophilic bacillus with which the disease could be reproduced in healthy chickens by inoculation (Fig. 13.1). This organism, to which the

name *Hemophilus gallinarum* (Eliot and Lewis, 1934) was given, is considered to be the primary cause of the disease now termed infectious coryza. In pathological exudates and in cultures 24 to 48 hours old, the most characteristic form is a polar staining rod. Beaded threads and filamentous forms are also seen in young cultures, but after 48 hours they have a tendency to undergo fragmentation or degeneration. The bacillus can be readily demonstrated microscopically in exudate and obtained in culture during the early stages of the disease. In older cases, however, it is difficult or impossible to
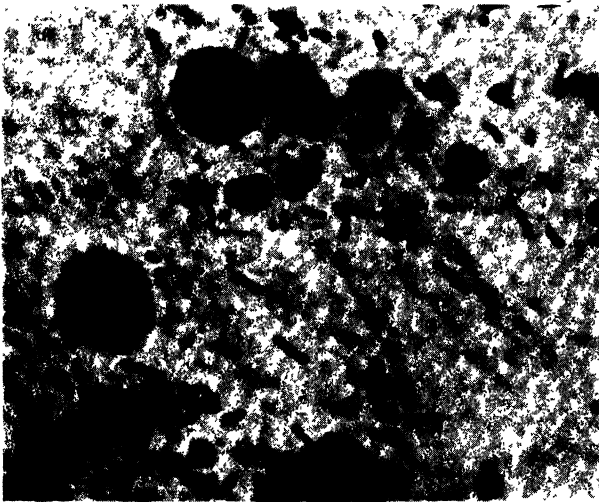


FIG. 13.1. *Hemophilus gallinarum* in film of nasal exudate. ×810.

identify because of the numerous other bacteria which are present. Consequently, to identify a field case as infectious coryza, it is often necessary to inoculate chickens with the nasal exudate in order to secure for examination a case which has just begun to show symptoms.

The coryza induced by inoculation with culture is usually of shorter duration than that from natural infection or that induced by inoculation with nasal exudate. This has raised the question of whether the natural disease is caused by *Hemophilus gallinarum* alone or in combination with another etiological agent.

Schalm and Beach (1936a) found that the culture-induced disease could be increased in both duration and severity by rapid serial passage through chickens. They also reported inability to demonstrate a second pathogenic agent, either bacteria or virus, in nasal exudate. They concluded, therefore, that the relative mildness of culture-induced disease was due to reduction in the virulence of the organism by cultivation in an artificial medium.

Nelson (1933b) referred to the culture-induced and exudate-induced disease as representing two types, the former of rapid onset and short duration, the latter of rapid onset and long duration. The hemophilic bacillus was regularly present with both types. He (Nelson, 1936a, 1936b, 1936c, 1936d) also reported a type of slow onset and long duration, the cause of which he found to be minute bodies, termed "cocobacilliform bodies," which he succeeded in cultivating in chicken embryos and in tissue culture. The

coryza bacillus was never present in this type of the disease. Later Nelson (1938) reported that the infection with both *H. gallinarum* and cocobacilliform bodies would produce a coryza of rapid onset and long duration, and offered this finding as an explanation of the difference in the duration of the culture-induced and exudate-induced disease.

Erwin, Delaplane, and Stuart (1937) have reported the isolation of still another organism, *Shigella nasalis* sp. nov., which is capable of acting in conjunction with *H. gallinarum* to increase the severity of the infection. This organism is described as a secondary invader which alone is incapable of causing coryza.

Gibbs (1935) reported having demonstrated that a filtrable virus was the cause of two outbreaks of epidemic colds in which hemophilic bacteria could not be isolated.

From the preceeding it is seen that the complete etiology of infectious coryza possibly has not yet been determined.

**Symptoms.** Infectious coryza has shown a remarkable variability in its severity and harmfulness to a flock. The mildest type of the disease occurs as a simple coryza with a nasal discharge, which may be persistent or of short duration, as the only symptom. Indications of systemic effect such as droopiness and diminished appetite are absent.

In the more severe types of the disease the coryza is complicated by other manifestations. One frequently seen is edema of the face which may extend, especially in males, to the intramandibular space and wattles (Figs. 13.2 and 13.3). Other complications are sinusitis, conjunctivitis, tracheitis, bronchitis, and infection of the air sacs. The severity of an outbreak is proportional to the character of the complications and the number of birds in which they are present. The heaviest losses occur when the incidence of lower respiratory tract involvement is high; in such cases coughing and gasping are prominent symptoms, and there are numerous deaths from suffocation which gives the disease a close resemblance to laryngotracheitis. In addition, the fowl are depressed, have diminished appetite, and become progressively emaciated.

The course of infectious coryza is usually prolonged, extending over a period of several weeks or months. The resultant mortality varies from a negligible number to more than 50 per cent of a flock. In an extreme case the disease persisted in a flock of 2,500 pullets from May to December and ultimately caused the destruction of the entire flock. In many outbreaks, however, the loss from decreased egg production and the development of worthless culls exceeds that due to immediate mortality. This seemingly harmless malady should not be ignored because its adverse effect on egg production may be considerable. It is likely to be recurrent in each year's replacement stock of young birds, and it may change to a severe type at any

time. Such a change in the character of coryza has been repeatedly brought about by the rapid passage of the infection from one chicken to another in the laboratory and has been observed on poultry farms.

**Species susceptible.** Pigeons have been found refractory to infectious coryza. The disease has been transmitted to turkeys, and the resultant disease was of the same character as natural cases of coryza and sinusitis in this species of bird. *H. gallinarum,* however, has not been identified in natural cases of sinusitis in turkeys.



FIG. 13.2. Artificial infection with infectious coryza showing facial edema.

**Transmissibility.** Infectious coryza has been transmitted experimentally by indirect contact, but it did not take place readily. It is easily transmitted, however, by confining healthy chickens with infected ones in cages or small pens and by the instillation of exudative material from any infected part into the nasal passages of susceptible chickens. The usual mode of transmission on a farm is probably direct contact between healthy chickens and those with active or latent infection. The importance of attendants as spreaders of the infection about a farm is unknown although trapnesters have been incriminated as probably having carried the disease from affected to healthy pens of breeding birds.

**Diagnosis.** The definite identification of a respiratory disease as infectious coryza requires the demonstration that the disease is transmissible and that *H. gallinarum* is present. The former can be accomplished without special facilities simply by the intranasal inoculation of healthy chickens from a coryza-free source with nasal exudate of an affected bird. For this purpose a bird in the early stages of the disease should be selected. Laboratory facilities are necessary for demonstrating the presence of the causative organism. Ordinarily it is preferable to submit specimens to a laboratory where both transmission trials and bacteriological procedures can be done. Experience has shown that any respiratory disease which has a nasal discharge as a constant symptom, affects several chickens at a time, persists in a flock over a period of weeks or months, and especially one which affects the pullets year after year is likely to be infectious coryza. It is desirable to have a field diagnosis of a disease manifested by coryza alone or in combination with edematous swelling of the face or wattles checked by a laboratory examination because identical symptoms can result from localized fowl cholera

infection. When the coryza is accompanied by symptoms indicative of tracheal or bronchial involvement; i.e., coughing and gasping, laboratory diagnostic procedures are necessary to determine whether the syndrome is due to coryza alone or to concurrent infection with coryza and infectious laryngotracheitis, infectious bronchitis, or avian pneumoencephalitis (Newcastle disease). In some instances when a slight nasal discharge is the only symptom, the possibility that it is the result of a borderline vitamin A deficiency in the diet should be considered.

**Prevention.** As stated previously, direct contact between infected and healthy chickens is the usual means by which infectious coryza is disseminated. In this case, both actively diseased chickens and those which have recovered from the disease and have become "healthy carriers" of the causative agent would be classed as infected chickens. Prevention, for the most part, consists of avoiding all contact between noninfected individual birds and flocks and those which are or have been infected.

The most effective single precaution to avoid introduction of the



FIG. 13.3. Edema of the wattles following intranasal injection of *Hemophilus gallinarum.*

disease into a clean flock is to make all replacements or additions with day-old chicks or hatching eggs. This applies both to the replacements which are routinely made each year and to the stock, including young males, which are secured for breeding purposes. Other measures of hygiene and sanitation designed to prevent the introduction of infectious agents by indirect means should also be observed. Prevention is more difficult in a congested poultry district in which the disease is prevalent, but it can be accomplished if the chickens are confined in houses without yards or with small well-fenced yards, which are located a reasonable distance from those on neighboring farms, and if any stray chicken, regardless of source, which is found outside the enclosure, is immediately destroyed.

The prevention of recurrences of the disease after it has once appeared on a farm requires complete and permanent separation of the survivors in infected groups from all other chickens either by means of segregation or depopulation. The latter has been the more effective of the two procedures.

In segregation the infected portion of the flock should be confined in a house as far removed from others as possible and be kept thus separated as

long as any of the birds remain. The caretaker should not be required to tend other chickens, but if this is necessary the work should be organized so that the infected house is the last to receive attention. A separate set of utensils should be provided.

Procedures for depopulation which have been successfully used are as follows: (1) The condemned flock was sold as market poultry and the farm restocked with day-old chicks; (2) the condemned flock was moved to other premises, usually rented for the purpose, and replaced with day-old chicks; the condemned flock was maintained in its new location until all of the birds were disposed of in the customary manner; (3) the replacement chicks were reared, until about ready to lay, where they were not exposed to coryza. The condemned flock was then sold as market poultry and replaced by the healthy pullets. In procedures (1) and (2) the replacement chicks may be on hand as long as two months before the condemned flock is removed, if the brooder houses are well separated from other poultry, and separate attendants and utensils are provided for the chicks. The replacement and condemned stock being maintained on different farms under procedures (2) or (3) should have separate attendants and utensils. Poultrymen who have succeeded in eliminating coryza by depopulation regard it as practical and much less costly than the loss by death, retarded development of the birds, and reduced egg yield which results when each year's crop of young birds becomes affected with infectious coryza.

Avian mixed bacterin has not proved effective for the prevention of coryza.

**Treatment and control.** Delaplane and Stuart (1941), using chickens artificially infected with cultures of *Hemophilus gallinarum,* demonstrated that sulfathiazole was capable of preventing the development of clinical symptoms when medication was started before the birds were inoculated, and of hastening the recovery of birds treated after symptoms had developed. The drug was said to be effective either when given in measured doses to individuals or when mixed with the mash. Similar findings were made by Hamilton (1943) and Beach (1943) in experimental treatment respectively of field cases of coryza and infection induced by inoculation with exudative material from field cases. Later Wernicoff and Goldhaft (1944) reported that the treatment offered an effective, practical, and not too costly means of controlling acute outbreaks of infectious coryza. Feeding dry mash medicated with 0.5 per cent sulfathiazole proved to be a practical and effective flock treatment for controlling outbreaks of acute infectious coryza in farm flocks and also for keeping the disease in check until such time as the infected flock might be disposed of as an eradication measure. In all of the work cited above, it was noted that both the artificially infected birds in which the development of symptoms had been prevented by sulfathiazole

therapy and the birds with clinical symptoms which appeared to have been cured by the drug were likely again to develop symptoms of coryza soon after the cessation of treatment. Furthermore, recurrence of the disease has been frequently observed in treated flocks since sulfathiazole therapy for coryza has come into more common use. Sulfathiazole is considered a valuable agent for curtailing loss from acute infectious coryza, for preventing the rapid spread of the disease through a flock, for treatment of individual birds, and for special uses such as the protection of breeding males upon their introduction into a flock where they will be exposed to the infection. Sulfathiazole therapy, however, cannot be relied upon to effect permanent cures and prevent recovered birds from remaining carriers of the infection. The use of the drug should not be regarded as replacing sanitary measures as a means of eradicating the disease on a farm.

The usual method for the administration of sulfathiazole is as 0.5 per cent of a dry mash. Wernicoff and Goldhaft (1944) state that moist mash containing the drug is not readily consumed by the chickens. The medicated mash is fed until nearly all of the affected birds are free from symptoms. This usually should not require more than 3 to 5 days. The treatment can be continued safely for a longer period, but if definite improvement in the condition of the flock is not seen within 7 to 10 days, continuance is unlikely to be beneficial. If, as is frequently the case with severely affected flocks, the appetite of the birds is depressed, the sulfathiazole content may be increased to 1.0 per cent at the start of treatment and reduced to 0.5 per cent as the food intake increases. Individual birds may be treated by giving 0.25 gm. to 0.5 gm. of sulfathiazole three or four times daily for 2 or 3 days. The most satisfactory results from flock treatment with the medicated mash can be expected when treatment is begun in the initial stage of an outbreak. The treatment can be repeated whenever the disease recurs, or for 1 or 2 days at intervals of 10 or more days as á precautionary measure. Certain pharmaceutical manufacturers recommend the use of sodium sulfathiazole dissolved in the drinking water at the rate of 1:1,000 instead of medicated mash. Delaplane and Stuart (1941) and Wernicoff and Goldhaft (1944) regard the use of sulfathiazole to be of diagnostic value because treatment with it will not be effective unless *Hemophilus gallinarum* is the causative agent of the respiratory disease present in the flock.

In outbreaks of infectious coryza some benefit may be derived from the removal of affected birds from the flock. The birds removed should be either permanently isolated or eliminated by sale or slaughter. The birds isolated may be treated with sulfathiazole and also, if desired, the flock from which they were removed. If this procedure is begun early and conscientiously continued, the spread of the infection may be so retarded and the severity so reduced that the disease will cause small loss and may be eradicated. Little

benefit can be expected from this procedure, however, unless it is started before many of the birds have become infected.

When the appetite of a flock is impaired, an endeavor should be made to stimulate food consumption by modifying the feeding practice.

It is always advisable to search for possible contributing factors such as faulty housing conditions or diet and intestinal parasitism which may influence the seriousness of the disease and retard recovery.

## REFERENCES

Beach, J. R.: 1943. Unpublished data.

——— and Schalm, O. W.: 1936. Studies of the clinical manifestations and transmissibility of infectious coryza of chickens. Poultry Sci. 15:466.

de Blieck, L.: 1932. A hemoglobinophilic bacterium as the cause of contagious catarrh of the fowl. Vet. Jour. (London) 88:9.

———: 1934. *Coryza infectiosa gallinarum.* Proc. Twelfth Internat. Vet. Cong. 3:161.

Delaplane, J. P.: 1936. The isolation of a hemophilic bacillus in pure culture and the reaction of chickens to extranasal inoculation thereof. Jour. Agr. Res. (U.S.A.) 52:377.

———: 1942. Suggestions for the use of sulfathiazole in the prevention and treatment of infectious coryza (*H. gallinarum* infection). Lederle Bul. 11:27.

———, Erwin, L. E., and Stuart, H. O.: 1934. A hemophilic bacillus as the cause of an infectious rhinitis. R. I. Agr. Exper. Sta., Bul. 244:1.

——— and Stuart, H. O.: 1941. The chemotherapeutic value of sulfathiazole in preventing and treating infectious coryza (*Hemophilus gallinarum* infection) in chickens. Jour. Am. Vet. Med. Assn. 99:41.

Eliot, C. P., and Lewis, M. R.: 1934. A hemophilic bacillus as a cause of infectious coryza in the fowl. Jour. Am. Vet. Med. Assn. 84:878.

Erwin, L. E., Delaplane, J. P., and Stuart, H. O.: 1937. *Shigella nasalis,* sp. nov., a secondary invader of the nasal passages of chickens. Jour. Am. Vet. Med. Assn. 91:317.

Gibbs, C. S.: 1935. The etiology of epidemic colds in chickens. Science 81:345.

Hamilton, C. M.: 1943. Treatment of infectious coryza in chickens with sulfathiazole. Jour. Am. Vet. Med. Assn. 103:141.

Nelson, J. B.: 1932. Etiology of an uncomplicated coryza in the domestic fowl. Proc. Soc. Exper. Biol. and Med. 30:306.

———: Studies on an uncomplicated coryza in the domestic fowl:

1933a. I. Isolation of a bacillus which produces a nasal discharge. Jour. Exper. Med. 58:289.

1933b. II. The relation of the bacillary coryza to that produced by exudate. Jour. Exper. Med. 58:297.

1936a. V. A coryza of slow onset. Jour. Exper. Med. 63:509.

1936b. VI. Coccobacilliform bodies in birds infected with coryza of slow onset. Jour. Exper. Med. 63:515.

1936c. VII. Cultivation of the coccobacilliform bodies in fertile eggs and in tissue culture. Jour. Exper. Med. 64:719.

1936d. VIII. The infectivity of fetal membrane and tissue culture suspensions of the coccobacilliform bodies. Jour. Exper. Med. 64:759.

1938. IX. The cooperative action of *Hemophilus gallinarum* and the coccobacilliform bodies in the coryza of rapid onset and long duration. Jour. Exper. Med. 67:847.

Schalm, O. W., and Beach, J. R.: 1934. The etiology of a respiratory disease of chickens. Science 79:416.

———: 1936a. Cultural requirements of the fowl coryza bacillus. Jour. Bact. 31:161.

———: 1936b. Studies of infectious coryza of chickens with special reference to its etiology. Poultry Sci. 15:473.

Wernicoff, N. E., and Goldhaft, T. M.: 1944. The field use of sulfathiazole in some diseases of poultry. Cornell Vet. 34:199.

# BRUCELLOSIS, ANTHRAX, PSEUDOTUBERCULOSIS, TETANUS, AND VIBRIO INFECTION

By H. J. STAFSETH, *Department of Bacteriology and Hygiene,*
*Michigan State College, East Lansing, Michigan*

☆   ☆   ☆

## BRUCELLOSIS

Brucellosis is the name applied to the disease caused by the three species of the genus Brucella. In the initial stages of the infection there may be a bacteriemia. After the bacteria recede from the blood stream, the disease continues in a subacute or chronic form with or without outward manifestations.

**Occurrence.** This disease is common in most countries in cattle, goats, swine, and man, and it also affects other mammals. It does not seem to be prevalent in birds. Veterinary literature contains several reports dealing with natural and experimental infection in various avian species. However, most of the diagnoses of brucellosis in fowl have been based on the agglutination test or merely on the fact that the sick birds were or had been in contact with infected mammals. Very rarely has any of the Brucella been isolated from naturally infected birds. Therefore, the possibility exists that the outbreaks of disease diagnosed as brucellosis might have been caused by something else. Furthermore, it is possible that the positive agglutination tests obtained were not due to established Brucella infection but to the picking up of material, contaminated with Brucella, from the ground, litter, feed, etc., in quantities sufficient for antigenic effect.

**Historical.** According to several authors brucellosis was first observed by Fiorentini in Italy in 1906. His diagnosis was based on the fact that 55 per cent of the birds reacted positively to the *Br. abortus* agglutination test. Dubois (1910) reported on an outbreak of a disease in chickens, with a 75 per cent mortality, which he regarded as brucellosis because the birds were in contact with sheep suffering from *Br. melitensis* infection, and 60 per cent of the affected birds reacted to the melitensis agglutination test in dilutions of 1:50 to 1:600. Zwick and Zeller (1913) unsuccessfully attempted to infect fowl by repeated subcutaneous, intramuscular, intraperitoneal, and intravenous injections of *Br. abortus*. Koegel (1923) tried to produce infection in chickens and pigeons by feeding and injecting large doses of *Br. abortus*.

These birds gave agglutination reactions in titers of 1:200 to 1:1,000 but showed no symptoms. Emmel and Huddleson (1929) and Huddleson and Emmel (1929) claimed to have produced infection in fowl by feeding naturally infected milk, portions of an aborted fetus, and cultures. They also claimed to have found natural Brucella infection in four flocks. A year later Emmel (1930b) found that turkeys, pheasants, ducks, and geese could be infected by feeding massive doses of the organism. During the same year he (Emmel, 1930a) found 16.5 per cent reactors to the agglutination test in a flock of ninety chickens. Many of these birds were in poor condition, and the egg production was low. According to Anguelov (1931) brucellosis is very prevalent, especially in modern poultry plants in Bulgaria. McNutt and Purwin (1930a, 1930b) tested 20 flocks with the agglutination test and found only a small percentage of reactors, none of them showing symptoms. Attempts at producing infection by feeding and injection failed, as no symptoms were shown and no deaths occurred. They further tested 10,000 birds in sixty-nine flocks and obtained less than 2 per cent reactors, the highest percentage in any one flock being 12. Here again no evidence of illness was observed. Strange and Beach (1931) failed to produce clinical disease in thirty-two chickens by feeding and injection of cultures. Gilman and Brunett (1930) tested four flocks of chickens, finding only a small percentage of reactors to the agglutination test. They conclude, however, that the evidence points to the presence of natural infection in farm flocks. McNutt and Purwin (1932) found laying pullets susceptible to infection with Brucella to the extent that egg production was slightly decreased temporarily. In 10-day chicks no ill effects were produced.

Van Roekel and his co-workers (1932) tested 25,202 chickens in fifty-three flocks with the agglutination test. The area covered represented approximately every county in Massachusetts. The total chicken population in these flocks was 70,479 birds. Two dilutions (1:25 and 1:50) were used. No reactors were found. Thirty flock owners had other livestock on the premises, and nine permitted the chickens to come in contact with the livestock. On three farms where reactors were found among cattle, the chickens were not allowed to come in contact with the cattle.

Two hens were given twelve feedings of a saline suspension of the organism during a period of 17 days, and agglutinins were detected 16 days after the first feeding. However, at no time were agglutinins well established in the blood stream, and the titer began to decrease two weeks after agglutinins were first detected. The birds were killed 28 days after the first feeding. Pathological and bacteriological findings were negative. Two hens were given three intraperitoneal inoculations with a saline suspension of the organism. Agglutinins were detected 7 days after the first inoculation, and a strong titer had been established at four weeks. At the end of seven weeks the

birds were killed, and no pathological and bacteriological evidence of infection was observed. Two males were given nine intraperitoneal inoculations with a saline suspension of the organism. Inappetence, somnolence, ruffled feathers, and decrease in body weight were observed. Agglutinins were present in the blood stream 9 days after the first feeding. Necropsies were performed 20 days after the first feeding, and *Br. abortus* was recovered from both individuals. One male was given three intravenous inoculations with a saline suspension of the organism. Clinical symptoms were observed. Agglutinins were established in the blood stream between the fourth and nineteenth days after the first inoculation. A necropsy was performed on the nineteenth day and *Br. abortus* recovered. These observations show that natural Brucella infection in chickens in Massachusetts appears to be of little, if any, significance. Agglutinins were produced when birds were fed and inoculated with saline suspensions of the organism. Repeated doses of the antigen were tolerated without producing death.

Beller and Stockmayer (1933) showed that chickens can be infected by natural and artificial channels. They found that the agglutination titer may be high 1½ years after infection. The organisms disappeared rapidly from the blood and organs and were not transmitted to the eggs. Young chickens were relatively resistant to the disease. In chickens injected with the organisms, agglutination titers as high as 1:10,000 were reached. The organisms were re-isolated from twelve of thirty-two chickens. Five of these were injected intravenously, six intramuscularly, and one intraperitoneally. In most cases the organisms were obtained from the spleen, liver, and bone marrow. Thomsen (1934) exposed 2,677 chickens to natural infection. Only 15 per cent of his birds developed agglutination titers of 1:50. He concluded that brucellosis in chickens is of no importance in Denmark. Liddo (1934) exposed birds to infection with *Br. melitensis* of human origin and found pigeons more susceptible than chickens. Pavlov (1938) reported on his experiments with brucellosis in birds. He found that birds are less easily infected than mammals by natural and artificial methods, and that chickens are more susceptible than pigeons. Five of seven rabbits placed with infected chickens became infected and died within three months. Pure cultures of Brucella were isolated from them. None of three normal chickens and ten normal guinea pigs, placed with infected chickens, developed any evidence of infection. Eggs from chickens injected with massive doses of Brucella were found to contain the organism only between the fourth and the fourteenth day after the injection. None of the injected birds developed any symptoms. The allergic and the agglutination tests were found to be effective procedures in the detection of infection. The former gives the most pronounced reaction 24 hours after the injection of the test substance and is applicable three months after infection. The agglutination titer reached its maximum one

month after infection. Two months after infection the agglutinins disappeared from the blood. Brucella, injected in small doses (0.01 cc.) into eggs kept at ordinary temperature, retained their virulence for one month and perhaps longer. Pagnini (1939) attempted to infect chickens by giving them gelatin capsules containing Brucella. The purpose of his work was to determine the role of chickens in the spread of brucellosis. Since he succeeded only when employing huge quantities of organisms, he concluded that chickens are of no practical significance in the spread of this disease.

It is evident that the results recorded by these authors are somewhat contradictory. One might account for the difference in results on the basis of variability in the virulence of the organisms and the resistance of the hosts employed in the various experiments. Generally considered, it seems that brucellosis in birds cannot be of much importance.

**Spread.** Contact infection among birds does not seem to take place. However, the fact that infected birds conveyed infection to rabbits shows that the organisms must have been passed with the droppings. The seat of the infection may have been in the digestive tract or in the urinary organs. Infected birds, eaten by other birds or by hogs, would, no doubt, serve as sources of infection.

**Symptoms.** Most authors have failed to observe symptoms of brucellosis in birds, and it is not certain that the symptoms described by others have actually been due to this disease. Dubois (1910) attributed the following symptoms to brucellosis: Between the months of March and June, 1910, 140 of 200 birds died. Young as well as old birds were affected. The disease appeared in a peracute and subacute form, killing birds in a few hours to 8 to 10 days. In the peracute form the birds died without previous symptoms. In the subacute form there was first loss of appetite; then the birds became weak and walked with difficulty. During the last 3 to 4 days they squatted and crowded together with ruffled feathers and drooping wings; they could easily be caught; sometimes they had greenish diarrhea. At last they became extremely emaciated. The following symptoms were recorded by Emmel and Huddleson (1929) : "The birds first went off egg production and usually developed severe diarrhea. A gradually increasing paleness about the head, comb, and wattles, and emaciation occurred. Before death the birds became weak and often showed paralysis. The course of the disease ranged from 18 to 46 days."

**Pathology.** Dubois (1910) observed swelling of the spleen and liver and petechiae on the lungs. Emmel and Huddleson (1929) recorded the following anatomical changes: In the early stages of the disease the spleen was generally enlarged; later in the course of the disease it became shrunken. The liver was pale, mottled, and showed numerous brownish or gray foci on the surface; at death it had become pale and friable. The kidneys were also

pale and showed some degeneration. The ovary was in the process of atrophy with flaccid ova of a dirty yellowish color. The intestines showed inflammation with some necrosis. In the duodenum there were irregular, elevated areas of cell infiltration. Beller and Stockmayer (1933) mention enlargement of the liver and very rare occurrence of necrotic foci in the spleen.

**Diagnosis.** The isolation and identification of one of the members of the genus Brucella constitutes the only positive way of diagnosing this disease. To be sure, agglutination and allergic tests have been employed successfully in experimental work. However, positive tests may be obtained with birds that cannot be proved to be infected by bacteriological examination, and Emmel and Huddleson (1929) found that the birds often gave a negative agglutination test in the last stages of the disease. Pavlov (1938) found that the agglutinins disappeared from the blood two months after infection, while Beller and Stockmayer (1933) found agglutinins in the blood a year and a half after infection. The difference between the observations of Pavlov and Beller and Stockmayer may be due to a difference in the duration of the infection. According to Pavlov the allergic state remains longer than do the agglutinins in the blood. The allergic test was made by injecting a test substance, prepared according to Dubois (1933), into the skin at the point of the wing after removal of a few feathers.

**Treatment and prevention.** There is no treatment available for brucellosis in birds. Since birds can become infected with Brucella and may thus serve as agents of transmission of this disease, not only to other birds but to mammals as well, one should take steps to prevent fowl from being in contact with infected mammals. Good poultry hygiene demands that poultry should be confined within premises set aside for this type of livestock and not be allowed in barns, hog yards, etc. The practice of throwing dead chickens on the manure pile or elsewhere, where hogs or other birds may eat them, is to be condemned. Should a farm flock be found to be affected with brucellosis the safest thing to do will be to destroy the flock, disinfect the premises as carefully as possible and restock after leaving the poultry house and yards idle for several months. It has not been shown that the testing and slaughter method is applicable in poultry practice.

### REFERENCES

Anguelov, S.: 1931. Bul. de l'Office Internat. des Epizooties.
Beller, K., and Stockmayer, W.: 1933. Die Pathogenität der *Br. abortus* für Hühner und Kücken. Deutsch. tierärztl. Wochenschr. 41:551.
Dubois, C.: 1933. Dépistage des *Brucella* chez la poule par la récherche des réactions d'allergies. Compt. rend. Soc. de biol. 113:1045.
Dubois, M.: 1910. Malta fever in fowls. Rev. Vet. 67:490.
Emmel, M. W.: 1930a. An outbreak of Brucella disease in the fowl. Jour. Am. Vet. Med. Assn. 76:564.
———: 1930b. The susceptibility of the turkey, pigeon, pheasant, duck, and goose to Brucella disease. Jour. Am. Vet. Med. Assn. 77:185.
——— and Huddleson, I. F.: 1929. Abortion disease in the fowl. Jour. Am. Vet. Med. Assn. 75:578.

Gilman, H. L., and Brunett, E. L.: 1930. *Bact. abortus* infection in the fowl. Ann. Rep. N. Y. St. Vet. Coll., 1929–30, p. 109.

Huddleson, I. F., and Emmel, M. W.: 1929. The pathogenicity of the species of the genus Brucella for the fowl. Mich. Agr. Exper. Sta., Tech. Bul. 103.

Liddo, S.: 1934. Brucellosi aviaria sperimentale. Bol. Accad. Pugliese Sci., p. 100.

Koegel, A.: 1923. Beiträge zur Abortusforschung. Münchener tierärztl. Wochenschr. 74:617.

McNutt, S. H., and Purwin, P.: 1930a. The effect of the Brucella group of microorganisms on chickens. Jour. Am. Vet. Med. Assn. 77:212.

———— and Purwin, P.: 1930b. The effect of the Brucella group of microorganisms on chickens. Jour. Am. Vet. Med. Assn. 77:350.

———— and Purwin, P.: 1932. Feeding of Brucella organisms to chickens and its effect on egg production of pullets and on growth of young chicks. Jour. Am. Vet. Med. Assn. 81:641.

Pagnini, Ugo.: 1939. I polli nella diffusione delle brucellosi. La Clin. Vet. 62:75.

Pavlov, P.: 1938. La brucellose chez les volailles. Rec. de Méd. Vét. 114:790.

Strange, C. R., and Beach, B. A.: 1931. Are chickens susceptible to contagious abortion? Vet. Med. 26:4.

Thomsen, A.: 1934. Sur la présence de l'infection à Brucella dans les effectifs de Volaille du Danemark. Bul. de l'Office Internat. des Epizooties. 7:1037.

Van Roekel, H., Bullis, K. L., Flint, O. S., and Clarke, M. K.: 1932. Susceptibility of chickens to brucelliasis. Jour. Am. Vet. Med. Assn. 80:641.

Zwick and Zeller: 1913. Über den infektiösen Abortus des Rindes. Arb. a.d. kaiserl. Gesundheitsamt. 43:1.

## ANTHRAX

Anthrax is caused by *Bacillus anthracis* which in susceptible hosts produces an acute septicemic (bacteriemic) infection characterized mainly by fever, slight coagulation and dark discoloration of the blood, swelling of the spleen, and edema and hemorrhages in various tissues.

**Historical.** Anthrax occurs rarely in birds. The rather frequent reports on cases of avian anthrax, found in the literature of the nineteenth century, were not based on bacteriological examinations, and it seems reasonable to assume that the great losses recorded may have been due to cholera or plague. The opinion expressed by Pasteur (1878) that chickens under ordinary conditions are resistant to anthrax, is shared by nearly all authors. Heusinger (1850) claimed that fowl could be fatally infected with anthrax. He is also responsible for the supposition that this disease occurred in epidemic form among chickens in Germany during the years 1832 and 1835.

### EXPERIMENTAL WORK WITH AVIAN ANTHRAX

A considerable amount of experimental work has been done on artificial infection and on the mechanism concerned in susceptibility and resistance.

**Artificial infection.** Feser (1879) injected twenty-four chickens subcutaneously, and gave others anthrax bacilli in their feed for weeks without producing a single case of anthrax. Renault fed anthrax material to birds without producing fatal infection. Brauell and Davaine fed anthrax infected meat to chickens with negative results. Oemler (1879) fed anthrax material to eight ducks and twenty-eight chickens. The ducks sickened in 24 hours and soon died, while none of the chickens became ill. Only negative results were obtained by Koch, Gaffky, and Loeffler (1884) who gave their chickens large numbers of spores in feed. Perroncito (1885), Kitt (1886), and Hess (1887) were unsuccessful in their attempts at producing anthrax in chickens.

In extensive experiments with anthrax in chickens, Hofherr (1910) failed to produce the disease even when giving large quantities of anthrax material to birds that were greatly weakened by starvation, thirst, cold baths, feeding of powdered glass, lime, vegetable diet, etc. By feeding a very large quantity of infectious material, one rooster, that was already sick, and one chick were infected. Fatal infection was produced by feeding in 25 per cent of the ducks and 100 per cent of the pigeons employed in his experiments. Thus, Hofherr showed that pigeons and ducks are susceptible to anthrax and that healthy chickens are ordinarily resistant but not always completely immune. Hunger and youth predispose to infection and so does lowered body temperature as was shown by Pasteur in 1878.

**Mechanism of resistance.** The mechanism of resistance of chickens to anthrax infection was studied very carefully by Wagner (1890). His work on forty-six chickens showed that under ordinary conditions chickens are resistant to this disease and that the immunity is due to phagocytic action of leukocytes. Anthrax bacilli can grow and maintain their virulence in the body of the chicken. The presence of the organism in the tissues is not without effect as shown by fever and cellular infiltration at the point of local infection. The resistance of chickens can be broken down by placing them in cold waterbaths for several hours or by giving them antipyrin or chloral hydrate. Submerging the chickens in a waterbath for several hours resulted in death from anthrax in all of six birds. Antipyrin lowered the resistance to the extent that six out of eleven birds died of anthrax. In the case of chloral hydrate only one bird out of eight died. Lowering of the body temperature was shown to interfere with the migration of leukocytes to the point of infection. The reason for the cold water bath being more effective in reducing resistance than were the other two agents used, was found in the fact that by it (the water bath) the temperature could be brought to a lower level and could be held there for a longer period than with antipyrin or chloral hydrate. Möllhoff (1910) tried to determine the reason for the great resistance of birds to anthrax, and on the basis of his experiments, drew the conclusion that the bactericidal action of lymph and blood play an important part, and that the high body temperature of the birds is less important.

## NATURAL INFECTION

**Anthrax in the ostrich.** Epidemics of anthrax have been observed in the ostrich. The disease usually runs an acute course, but subacute cases occur, often resulting in recovery. Anthrax in the ostrich was first reported by Henning in 1894. Robertson (1908) reported on a case in an ostrich, and later Theiler (1912), and others (1923) described the disease in detail.

**Pathology.** Anthrax bacilli are present in all organs and do not differ, even with respect to pathogenicity, from those isolated from mammals. The

following tissue changes may be observed: Slight coagulation and dark discoloration of the blood; increase in the fluid in the thoracic and abdominal cavities; petechial and larger hemorrhages in the pericardium, peritoneum and mesentery; sometimes gelatinous infiltrations in the subcutaneous and deeper tissues; very frequently there is hemorrhagic enteritis; edematous swelling and hemorrhages in many places in the submucosa; not infrequently there may be a mass of blood in the lumen of the colon; the spleen, liver, and kidneys show swelling and congestion with blood; as a rule the lungs and stomach appear normal.

**Anthrax in ducks.** In birds other than the ostrich, anthrax appears only sporadically. Gerlach (1923) described a case of natural infection in a duck. This case occurred shortly after a few pigs had died of anthrax on the same farm. Ubertini (1939) reported on an outbreak of anthrax in ducks. These ducks were kept with about 100 chickens. There were two varieties of ducks, the common kind and five "mute" ducks (*Cairina moschata*). These five became ill and died while none of the others nor the chickens were affected. This outbreak occurred about 10 days following the death of a cow from anthrax.

**Pathology.** The following tissue changes have been observed: edematous swelling of the head, throat, and upper part of the neck, sometimes extending along the entire length of the esophagus resembling a sac filled with fluid; the skin was bluish red in the affected areas; cyanosis of the mucosa of the head; gelatinous infiltration of the pharyngeal mucosa; inflammation of the intestinal mucosa and other changes such as those recorded in the following description by Almeyew of a case of anthrax in an eagle.

**Anthrax in birds in zoological gardens.** Almeyew (1936) reported that anthrax had been observed in zoological gardens following the feeding of meat containing large numbers of anthrax bacilli. The source of the meat was unknown. He gave detailed results of an autopsy performed by him on the carcass of an eagle that had died of anthrax. This eagle was sent in from the Kasaner Zoological Garden where it had died suddenly May 12, 1935.

The post-mortem findings were mainly as follows: The nutritional state of the carcass was fair; there was only slight muscular rigidity. The visible mucous membranes of the oral cavity showed a pale gray color, thick slimy coating, on the removal of which one could observe passive hyperemia. The tongue was covered with a slimy mass. The spleen was greatly enlarged; the capsule was distended, the pulp soft, discolored, and moist; in the pulp there were sharply circumscribed nodules of the size of pinheads to that of millet seed, reddish-brown or grayish-red in color; the cut surface was dull. The liver was enlarged, distended, and hyperemic; under the capsule and throughout the parenchyma there were hemorrhagic and necrotic foci. The cut surface was moist, and the vessels were distended. The kidneys were much

enlarged, hyperemic, spotted, grayish-yellowish brown in color and were studded with hemorrhages and necrotic foci. The cut surface was moist, and the exuding blood was only slightly coagulated; the cortical and medullary layers were hyperemic and studded with grayish-red foci. The adrenals were edematous, hyperemic, and showed numerous hemorrhages; the serous membrane was dull and showed petechial hemorrhages. The lungs were edematous, hyperemic, and dark bluish-red in color; the surface was moist and studded with petechial hemorrhages. The pericardium was distended, of dull bluish-red color and contained a yellowish-red transudate. The heart muscle was soft, and pale, loam-colored, and parboiled in appearance. The stomach was collapsed, grayish-blue; the vessels of the serosa were filled with blood; the stomach wall was thickened, the mucous membrane wrinkled, dark, and covered with mucus. The duodenum was dark red and showed numerous petechial hemorrhages and the mucous membrane was covered with a reddish-brown mucous mass. The surface of the large intestine was dark, brownish-red, the walls thickened and the mucous membrane covered with a brownish mucous layer.

**Diagnosis.** A diagnosis of anthrax in birds may be established by microscopic examination of stained films from blood, edematous fluid, or tissues in which anthrax bacilli usually are present in large numbers. It is well to confirm the diagnosis by cultural methods because of the possibility of mistaking saprophytic organisms for anthrax bacilli. The Ascoli precipitation test is a quick and reliable diagnostic procedure.

**Treatment and prevention.** Theiler (1912) recommends dilute carbolic acid. However, it is not to be expected that any medicinal treatment will be of value in treating birds for anthrax. Due to the rare occurrence of this disease in birds, preventive vaccination is not indicated. Furthermore, nothing is known as to the efficacy of this procedure as applied to birds. Every effort should be made to prevent the spread of infection. Sick birds should be removed promptly, killed, and burned. Under no circumstances must dead birds be allowed to lie around so that other birds may pick at them or eat them. Contaminated ground should be fenced off, and houses should be thoroughly cleaned and disinfected. Litter, rubbish, and equipment of little or no value, should be burned. Valuable birds that show no symptoms may be confined, preferably in houses with good cement floors where proper disinfection is possible. If the birds are few in number and of little value, it may be preferable to destroy the whole flock to facilitate the cleaning-up process.

## REFERENCES

Almeyew, H. S.: 1936. Anthrax beim Vogel. Deutsch. tierärztl. Wochenschr. 44:375.
Feser: 1879. Über Infektionsversuche mit Milzbrand beim Hausgeflügel. Adams Wochenschr. [Quotation from Kitt (p. 86).]
Gerlach, F.: 1923. Bemerkenswerter Verlauf einer Milzbrandenzootie. Wiener tierärztl. Monatschr. 10:481.

Hess, C.: 1887. Untersuchungen zur Phagocytenlehre. Virchow's Archiv. 109:365.

Heusinger, C. F.: 1850. Milzbrand Krankheiten der Thiere und des Menschen. Enke, Erlangen.

Hofherr, O.: 1910. Experimentelle Beiträge zur Milzbrandinfektion des Geflügels durch Fütterung. Zentralbl. f. Bakt. I. Orig. Bd. 55:434.

Kitt, T.: 1886. Kleinere Mittheilungen aus der pathologischen Abtheilung und Seuchenver-suchsstation. Jahresbr. der K. Central-Thierarzneischule in München (1884–85). Suppl. Deutsch. Zeitschr. f. Thiermed. u. vergleich. Pathol. XII:85.

Koch, R., Gaffky, G., and Loeffler, F. A. J.: 1884. Experimentelle Studien über die künstliche Abschwächung der Milzbrandbacillen und Milzbrandinfektion durch Fütterung. Mittheilung des Gesundheitsamt. 2:174.

Möllhoff: 1910. Untersuchungen über die Empfänglichkeit des Geflügels für Milzbrand und über die Gründe der Resistenz des Huhnes gegen diese Krankheit. Inaug. Dissert. Univ. Bern.

Oemler, H.: 1879. Experimentelle Beiträge zur Milzbrandfrage. Arch. f. wiss. u. prakt. Thierheilk. 5:164.

Pasteur, L.: 1878. Charbon et virulence. Bul. de l'Acad. de Med. 43:253.

Perroncito, E.: 1885. Carbonchio nei polli, p. 159: il Carbonchio, Mezzi preventivi e curativi. Torino. [Quotation from Kitt (p. 90).]

Robertson, W.: 1908. Case of anthrax in an ostrich. Jour. Comp. Path. and Therap. 21:361.

Theiler, A.: 1912. Anthrax in the ostrich. Agr. Jour. of Union of So. Africa 4:370.

Ubertini, B.: 1939. Un focolaio di infezione carbonchiosa ad insorgenza spontanea nelle anitre (Chairina Moschata). La Clinica Véterinaria 62:72.

Wagner, K. E.: 1890. Contribution à l'étude de l'immunité. Le Charbon des Poules. Ann. de l'Inst. Past. 4:570.

Ward, A. R., and Gallager, B. A.: 1923. Diseases of Domesticated Birds. The MacMillan Co., New York.

## PSEUDOTUBERCULOSIS

Pseudotuberculosis is a contagious disease caused by *Pasteurella pseudo-tuberculosis,* usually characterized by an acute septicemia of short duration, followed by a chronic focalized infection which gives rise to tubercular lesions in various organs.

**Historical.** The first isolation of the bacillus of pseudotuberculosis was made from a subcutaneous tubercular lesion on the forearm of a child by Malassez and Vignal (1883). They published another report on this organism in 1884. Rieck (1889) reported on an outbreak of a disease in canaries which, according to the description of the symptoms, lesions, and character-istics of the isolated organism, must have been pseudotuberculosis. Several other authors have reported on this disease in canaries, namely: Zürn (1884), v. Wasielewski and Hoffman (1903), Pfaff (1905), Freese (1907), Miessner and Schern (1908), Zwick (1908), Zeiss (1914), and van Heelsbergen (1927).

Woronoff and Sineff (1897) reported that they had found a case of pseudotuberculosis in a chicken. Truche and Isnard (1937) reported on an outbreak of this disease in a flock of adult chickens, and Schäfer (1939) reported on two cases in 14-day-old Leghorn chicks.

Bryner (1906) reported on an outbreak that occurred in 1904 involving tiger finches (*Habropya amandara* L., *H. melpoda* vieill), butterfly finches (*H. phoenicotis* sws.), and Japanese titmice. Pseudotuberculosis in pigeons was reported by Dolfen (1916) and by Lesbouyries (1934). Reports on pseudotuberculosis in turkeys have appeared by: Krage and Weisgerber (1924), Beck and Huck (1925), Lerche (1927a, 1927b), and Truche and

Bauche (1929). Christensen (1927) isolated *Bact. pseudotuberculosis rodentium* (Pfeiffer) from pseudotuberculous lesions of a variety of birds. Truche and Bauche (1933) have reported on an outbreak of pseudotuberculosis in fowls and pheasants, and according to Boquet (1937), they (Truche and Bauche, 1930) have also reported the disease in ducklings. Pseudotuberculosis in the swan was reported by Truche (1935). Urbain and Nouvel (1937) reported on the disease in toucans (*Rhamphastos cuvieri* Gould and *R. ariel* Vig.). Beaudette (1940) reported on a case of pseudotuberculosis in a blackbird. Bacteriological investigations were made in the years 1938 to 1945 by Karlsson (1945) of 80 cases of pseudotuberculosis in domestic animals, 13 of which were found in birds, including 10 cases in turkeys.

**Etiology.** The cause of this disease was first observed by Malassez and Vignal (1883) and was named *Bacterium pseudotuberculosis rodentium* by Pfeiffer (1890). Topley and Wilson (1936) and Bergey (1939) use the name *Pasteurella pseudotuberculosis.* In these two books and in the review of the literature on this disease by Beaudette (1940), there are complete descriptions of this organism. The earlier descriptions have been incomplete, making it difficult to identify newly isolated organisms on the basis of the available literature. Hutyra, Marek, and Manninger (1938) claim that the designation, *Pasteurella pseudotuberculosis*, is a misnomer. However, this name is used here in conformity with the nomenclature accepted by the Society of American Bacteriologists.

**Occurrence.** In the United States only one definitely identified case of avian pseudotuberculosis has been described, namely, the case in a blackbird in New Jersey reported by Beaudette (1940). There is one other report, a preliminary one by Kinyoun (1906), that may deal with this disease. This author encountered a disease resembling pseudotuberculosis in canaries coming from a dealer in Washington, D. C. Unfortunately, the description of the organism is too incomplete for identification. In Europe, particularly in Germany and France, the disease must be fairly common, canaries and turkeys being the most commonly and severely affected. It usually occurs as a sporadic disease in individual flocks. One must remember that pseudotuberculosis often affects wild rodents, field hares and rabbits, and also domesticated rabbits and guinea pigs. Thus, such animals may be the source of infection in birds if direct or indirect contact is permitted.

**Dissemination.** The usual avenue of infection is the digestive tract. Skin injuries may also form portals of entry. Predisposing causes are apparently important since, as a rule, the only birds affected are those whose resistance has been lowered by inadequate feeding, exposure to cold, and worm infestation. Very young birds are particularly susceptible. During cold and wet weather in the fall, considerable losses may occur among young turkeys.

**Pathogenesis.** In susceptible birds the organism gains entrance to the blood stream through breaks in the skin or through the mucous membranes, perhaps mostly (but not necessarily exclusively) in the digestive tract. Thus, a bacteriemia is established. Usually the bacteriemic condition is of short duration, but the bacteria are not all destroyed. Some of them establish foci of infection in one or more organs such as the liver, spleen, lungs, intestines, and kidneys, giving rise to tuberculous lesions. Such lesions have also been found in the mesentery and breast muscles.

**Symptoms.** The length of the incubation period of artificial infection varies considerably with the virulence of the organism, the amount of inoculum, the avenue of introduction, and the host species. Sparrows and canaries are very susceptible and may die in 1 to 3 days from small doses of organisms injected subcutaneuosly or intramuscularly. Feeding of cultures is usually ineffective unless some intestinal inflammation is present to act as a predisposing influence. Canaries, given mustard seed for the purpose of causing intestinal irritation, have sickened 5 days after being given cultures by mouth, death resulting 2 days later. Judging from the various reports available, it seems that the incubation period may be from 3 to 6 days in acute attacks and two or more weeks in chronic cases.

The symptoms also vary considerably. In very acute cases the birds may die suddenly without warning, or they may live a few hours or 2 to 3 days after showing the first symptoms. Such cases are usually marked by sudden appearance of diarrhea and the usual general manifestations of an acute septicemia. Usually, however, the course of this disease extends over two or more weeks, in which case the symptoms appear 2 to 4 days before death. In such cases the birds will show weakness, dull and ruffed feathers, and difficult breathing. Diarrhea is also a common symptom in such cases. Occasionally the disease will run a still more protracted course, when emaciation and extreme weakness or paralysis may be evident. Such manifestations as stiffness, difficulty in walking, droopiness, somnolence, constipation, and discoloration of the skin have also been observed. In the early stages of the chronic form of the disease, the birds may eat normally, but the appetite is usually completely lost 1 or 2 days before death.

**Anatomical changes.** In highly acute cases the only changes observed are swelling of the spleen and enteritis. Subacute or chronic cases result in enlargement of the liver, spleen, kidneys, and lungs. Yellowish-white foci of the size of millet seed may be found in the liver, spleen, lungs, kidneys, and breast muscles. There is usually severe enteritis, which is sometimes hemorrhagic. The serous cavities may sometimes contain more or less clear fluid.

**Prognosis and diagnosis.** The prognosis is unfavorable. A definite diagnosis can be established only by isolation and identification of the organism since the symptoms and lesions are very similar to those of several other

diseases, such as cholera, typhoid, paratyphoid, spirochetosis, tuberculosis, and certain forms of the leukosis complex. In making a bacteriological examination it must be remembered that the organism can be found in the blood in acute cases but must be sought in tissues in chronic ones.

**Treatment and prevention.** No medicinal treatment is available. Protective vaccination has not proved successful. Therefore, one must depend on the usual sanitary and hygienic procedures in combating this disease.

## REFERENCES

Beaudette. F. R.: 1940. A case of pseudotuberculosis in a blackbird. Jour. Am. Vet. Med. Assn. 97:151.

Beck, A., and Huck, W.: 1925. Enzootische Erkrankungen von Truthühnern und Kanarienvögeln durch hämorrhagische Septikämie (Paracholera). Zentralbl. f. Bakt. I. Orig. 95:330.

Bergey. D. H.: 1939. Manual of Determinative Bacteriology. 5th Ed. Williams and Wilkins Co., Baltimore. p. 294.

Boquet. P.: 1937. Recherches expérimentales sur la pseudo-tuberculose des rongeurs. Ann. de l'Inst. Pasteur 59:311.

Bryner, A.: 1906. Ein Beitrag zur Pseudotuberculose der Vögel. Inaug. Dissert. University of Zurich.

Christensen, N. P. C.: 1927. Pseudotuberkulose hos fugle foraarsaget af *Bacterium pseudotuberculosis rodentium* (Pfeiffer). Zentralbl. f. Bakt. I. Ref. 87:186.

Dolfen, H.: 1916. Über eine pseudotuberkulöse, seuchenhafte Erkrankung bei Tauben. Inaug. Dissert. Hannover.

Freese: 1907. Über seuchenhafte Erkrankung mit septikämischen Charakter bei Kanarienvögeln. Deutsch. tierärztl. Wochenschr. 15:501–5.

Hutyra, F., Marek, J., and Manninger, R.: 1938. Special Pathology and Therapeutics of the Diseases of Domestic Animals. 1:681–85. Alexander Eger, Chicago.

Karlsson, Karl-Fredrik: 1945. Pseudotuberkulos hos hönsfoglar. Skand. Vet. Tidskr. 35(11)673.

Kinyoun, J. J.: 1906. Bird plague. Science 23:217.

Krage and Weisgerber: 1924. Eine Putenseuche mit Diplo-Streptobazillenbefund. Tierärztl. Rundschau. 30:308.

Lerche: 1927a. Über eine neue Seuche der Truthühner. Arch. f. Geflügelk. 1:111.

————: 1927b. Die "Paracholera" der Puten und ihre Beziehung zur Pseudotuberkulose der Nagetiere. Zentralbl. f. Bakt. I. Orig. 104:493.

Lesbouyries, G.: 1934. Pseudo-tuberculose du pigeon. Bul. de l'Acad. Vet. de France 7:103.

Malassez, L.: 1884. Sur le microorganisme de la tuberculose zoogloéique. Arch. de Physologie Normale et Pathologique. Series 3, iv:81–105.

———— and Vignal, W.: 1883. Tuberculose Zoogloéique (forme ou especè de tuberculose sans bacilles). Arch. de Physiologie Normale et Pathologique. Series 3, ii:369–112.

Miessner and Schern: 1908. Die infektiöse Nekrose bei den Kanarienvögeln. Arch. f. wiss. u. prakt. Tierheilk. 34:133.

Pfaff, F.: 1905. Eine infektiöse Erkrankung der Kanarienvögel. Zentralbl. f. Bakt. I. Orig. 38:275.

Pfeiffer, A.: 1890. Über die bacilläre Pseudotuberculose bei Nagethieren. Zentralbl. f. Bakt. I. Orig. 7:219.

Rieck, M.: 1889. Eine infektiöse Erkrankung der Kanarienvögel. Deutsch. Zeitschr. f. Tiermed. u. Vergleich. Path. 15:68.

Schäfer. W.: 1939. Das Vorkommen des *B. pseudotuberculosis rod.* oder eines ihm ähnlichen Erregers bei Hühnerküken. Tierärztl. Rundschau 45:72.

Topley, W. W. C., and Wilson, G. S.: 1936. The Principles of Bacteriology and Immunity. 2nd Ed. William Wood and Co., Baltimore, pp. 607–9.

Truche, C.: 1935. Pseudo-tuberculose du cygne. Bul. de l'Acad. Vet. de France 8·278.

———— and Bauche, J.: 1929. La pseudo-tuberculose du dindon. Ann. de l'Inst. Pasteur 43:1081.

———— and Bauche, J.: 1930. Contribution a l'étude de la pseudo-tuberculose des oiseaux. Bul. de l'Acad. Vet. de France 3:391.

———— and Bauche, J.: 1933. Le bacille pseudotuberculeux chez la poule et la faisan. Bul. de l'Acad. Vet. de France 6:43.

Truche, G., and Isnard, S.: 1937. Un nouveau cas de pseudo-tuberculose chez la poule. Bul. de Acad. Vet. de France 10:38.

Urbain, A., and Nouvel, J.: 1937. Epidemie de pseudo-tuberculose chez des toucans de cuvier (*Rhamphastos cuvieri* Gould) et des toucans ariel (*Rhamphastos ariel* Vig.). Bul. de l'Acad. Vet. de France 10:188.

van Heelsbergen, T.: 1927. Pseudotuberculosis canariensis. Tijdschr. v. Diergeneesk. 54:545.

v. Wasielewski, and Hoffman, W.: 1903. Über eine seuchenhafte Erkrankung bei Singvögeln. Arch. f. Hyg. 47:44.

Woronoff, A., and Sineff, A.: 1897. Zur pathologischer Anatomie und Bakteriologie der bacillären Pseudotuberculose. Zentralbl. f. allg. Path. u. path. Anat. 8:622.

Zeiss, H.: 1914. Über einige bei Tierkrankheiten gefundene Erreger aus der Gruppe der hämorrhagischen Septikämie und der Koligruppe. Arch. f. Hyg. 84:1.

Zürn: 1884. Blätter f. Geflügelzucht. Dresden, p. 326.

Zwick, W.: 1908. Untersuchungen über eine Kanarienvögelseuche. Zeitschr. f. Infekt.-Krankh. d. Haust. 4:33.

## TETANUS

Tetanus is an acute infectious disease caused by the toxin of *Clostridium tetani* and is characterized by more or less persistent tonic spasms of some of the voluntary muscles resulting from an increased reflex irritability of the intoxicated motor nerve centers. The toxin is elaborated in infected wounds providing suitable conditions for growth and reproduction of the tetanus bacillus. Anaerobic wounds in which there is necrotic tissue or blood-effusion are particularly favorable.

**Occurrence.** Birds are comparatively very resistant to tetanus. Knorr (1899) has shown that the lethal dose of tetanus toxin for fowl per kilogram of body weight is 200,000 times greater than that for the horse. This fact no doubt accounts for the scarcity of reports dealing with tetanus in birds.

**Tetanus or tetanus-like disease in birds.** Dreymann (1894) reported on a case of illness resembling tetanus in a turkey. Four to 5 days after having been bitten by a dog, this bird showed a clumsy and stiff gait, the neck was extended, the neck muscles were hard and there was trismus. On the third day the entire body was rigid, the wings were held tightly against the body, the feathers were ruffled, the nictitating membranes and eyes protruded, there was almost complete trismus, and the bird died.

A case of tetanus in a young goose was described by Funck (1919). The symptoms were mainly trismus and difficult locomotion. The autopsy revealed a perforation of the stomach wall, caused by an iron wire, which in the author's opinion was the portal of entry.

Russell (1937), of Portsmouth, England, reported on a rapidly fatal case of illness in a young child, diagnosed as tetanus. An investigation revealed that the garden, in which the child played, was fertilized exclusively with pigeon manure. Tetanus bacilli were recovered from two of four soil samples, two samples of dirt or litter from the pigeon loft and from the excreta of one pigeon.

On July 15, 1940, Dr. L. H. Scamman of the Angell Memorial Hospital, Boston, Massachusetts, wrote me in part as follows: "It is a six-months-old domestic bird (goose) and has all the appearances of lock jaw. It cannot open its jaws at all, and if they are forced apart, they close by themselves."

Another case of a tetanus-like disease in a goose was reported to the

Michigan Agricultural Experiment Station, October 4, 1940. This goose was three months old and showed typical symptoms of tetanus.

In none of these four cases was there any bacteriological work done to prove the identity of the disease.

**Diagnosis.** A positive diagnosis of tetanus can be established only by the isolation and identification of the organism. This is not always possible due to the difficulty in locating the focus of infection and the fact that tetanus bacilli often soon disappear from infected wounds. Remedial effects obtained by the administration of tetanus antitoxin would constitute evidence of some diagnostic value.

**Treatment and prevention.** Antitoxin given in large doses may be of some value in the very early stages of the disease in mammals. There is no work showing the effect of antitoxic treatment in birds. The rare occurrence of tetanus in birds makes special preventive measures superfluous.

### REFERENCES

Dreymann: 1891. Zwei seltene Fälle von Tetanus beim Rind und Truthahn. Monatschr. f. Thierheilk. 5:75.
Funck, E.: 1919. Starrkrampf bei einer Gans. Tierärztl. Rundschau. 25:156.
Knorr, A.: 1899. Die Tetanuserkrankung und ihre Bekämpfung. Monatschr. f. Thierheilk. 10:211.
Russell, A. W.: 1937. Pigeons as possible tetanus carriers. Brit. Med. Jour. 2:1220.

## VIBRIO INFECTION

Vibrio infection is an acute infectious disease which resembles fowl cholera with respect to symptoms and gross pathology.

**Historical.** This disease was first reported by Gamaleia (1888) who observed it several times during the summer near Odessa. Krause and Windrath (1919) observed the disease in Germany in newly imported sunbirds (*Leiothrix luteus*). According to Hutyra, Marek, and Manninger (1938), Czukas has observed it in Hungary in fattened geese.

**Cause and dissemination.** The cause is *Vibrio metchnikovi* (Gamaleia), also called the paracholera vibrio. This organism is a small, bent, comma-shaped microbe that can be easily cultivated. It is very motile and is Gram-negative. Culturally and biochemically it resembles almost fully the vibrio of Asiatic cholera. These two organisms are also related antigenically as is shown by cross immunity. According to Czukas the organism infects birds that are weakened by poor hygienic conditions and by fattening. *Vibrio metchnikovi* is also pathogenic for pigeons and guinea pigs. Chicks can easily be infected per os. Older chickens and rabbits are very resistant. It appears that stagnant water and other materials contaminated with droppings of infected birds may serve as sources of infection.

**Symptoms.** In chickens the symptoms resemble those of cholera. Vibrio infection is generally less acute, and the body temperature is only slightly elevated while in fowl cholera it is usually high, e.g., 43°–44° C. The birds

observed by Gamaleia sat crowded together and disinterested, with ruffed feathers. Diarrhea was a constant symptom. The birds usually died in 2 to 3 days. In the sunbirds the disease was so acute as to cause the death of several hundred birds in a few days.

**Pathology.** The organism causes enteritis and gastro-enteritis which is sometimes hemorrhagic. It may enter the blood stream and give rise to necrotic processes in the liver and lungs. In young chickens and sunbirds the organism appears in the blood in large numbers, but in adult chickens the blood smears are mostly negative.

**Diagnosis.** A differential feature in the pathology of cholera and vibrio infection in chickens is that the latter produces no or few petechial hemorrhages in the intestinal tract and only slight or no changes in the organs. A definite diagnosis can be made only by the demonstration of the organism in the blood and preferably by the isolation and identification of the vibrio. Cultures should be made from the blood and organs showing lesions. The bacteriological diagnosis is complicated by the fact that the organism is usually confined to the lumen of the intestinal tract.

**Treatment and prevention.** No medicinal treatment is known, and prevention must depend on the usual sanitary measures employed in the control of diseases of similar epidemiological nature.

## REFERENCES

Gamaleia, N.: 1888. Sur l'étiologie du choléra des poules. Ann. de l'Inst. Pasteur 2:510.

Hutyra, F., Marek, J., and Manninger, R.: 1938. Vibrio cholera in fowls (Gastro-enteritis Cholerica Avium). Pathology and Therapeutics of the Diseases of Domestic Animals. Alexander Eger, Chicago 1:122.

Krause, W., and Windrath, H.: 1919. Über eine durch einen Vibrio veranlasste Seuche der Sonnenvögel (*Leiothrix luteus* L, chinesische Nachtigall). Berliner tierärztl. Wochenschr. 35:468.

# LISTERELLOSIS, BOTULISM, ERYSIPELOTHRIX, AND GOOSE INFLUENZA

*By* NORMAN D. LEVINE, *Department of Veterinary Pathology and Hygiene, College of Veterinary Medicine, University of Illinois, Urbana, Illinois*

☆ ☆ ☆

## AVIAN LISTERELLOSIS

A specific septicemia caused by the bacterium *Listerella monocytogenes* (*Bacterium monocytogenes, Listeria monocytogenes*) has been observed in chickens within recent years. Although the disease has as yet been reported by only a few workers, it is probable that it may be recognized more often in the future.

Listerella was first isolated by Murray, Webb, and Swann (1926) from an outbreak of disease in laboratory rabbits and guinea pigs in England. It was first found to be the cause of an encephalitis of sheep by Gill in 1931 in New Zealand, and has since been described from ruminants by a number of investigators. So far, Listerella has been found to be a cause of spontaneous disease in the rabbit, guinea pig, gerbille, sheep, cow, goat, fox, pig, chicken, horse, raccoon, capercailzie, and man. The literature on listerellosis up to 1943 is reviewed by Graham, Levine, and Morrill (1943).

Ten Broeck, in 1932, first isolated Listerella from chickens in the stock birds at Princeton University. This finding was recorded by Seastone (1935), who also reported on the characteristics of the disease and its causative organism. Paterson (1937) found Listerella to be the cause of death in outbreaks in four flocks of chickens in England, and later (1939) observed it in two other flocks. He stated that Watkins had isolated Listerella from twelve chickens in a number of flocks in England. Cole (1941) observed an outbreak of Listerella infection in the poultry flock at Cornell University in which twenty-six birds were affected. Hurt, Levine, and Graham (1941) isolated Listerella from a single chicken from a flock in Illinois. The organism was isolated from a chicken in California by Hoffman and Lenarz (1942). The first cases of listerellosis in chickens recognized in Germany were reported by Pallaske (1941), and Pothmann (1944) found the infection in chickens in East Prussia. Lilleengen (1942) cultured Listerella from a dead capercailzie hen (a large European grouse, *Tetrao urogallus*) at a game farm in Sweden.

**Etiology.** *Listerella monocytogenes* (Murray, Webb, and Swann, 1926; Pirie, 1940)[1] is a small Gram-positive rod (Fig. 15.1). Spores are not formed. It is motile, and when grown at 37° C. has a characteristic tumbling movement. This temperature is not so conducive to flagellum production, however, as is room temperature. The organism is a facultative aerobe. On nutrient and liver agar, circular smooth colonies are formed, which are bluish by transmitted light and milk-white by reflected light. After several days' growth on liver agar the colonies become very viscid. A clear zone of hemolysis is formed around colonies on blood agar. With some strains, hemolysis may not occur on initial isolation, but appears after several transfers. Gelatin is not liquefied; indol, acetyl methyl carbinol, and hydrogen sulfide are not formed; and nitrates are not reduced. In a study of the biochemical characteristics of 50 strains of Listerella, Harvey and Faber (1941) found that acid is produced from dextrose, levulose, galactose, maltose, mannose, rhamnose, trehalose, dextrin, and salicin; that lactose, glycerol, sucrose, arabinose, xylose, mannitol, sorbitol, and starch may or may not be fermented; and that acid is not produced from dulcitol, inositol, raffinose, glycogen, and inulin. In the writer's experience, sucrose may not be fermented on first isolation, but after several transfers on artificial culture media, this sugar is usually readily fermented. Dextrose, rhamnose, and salicin are usually fermented rapidly, while the other carbohydrates are usually fermented more slowly.



Fig. 15.1. *Listerella monocytogenes* from chicken. Gram stain. ×990.

Although it may grow quite lightly on initial isolation, after several transplants Listerella grows rather heavily both at 37° C. and at room temperature.

Several serologic studies have been carried out on Listerella. Using the agglutination test, Paterson (1940) recognized four serologic types, while Drew (1946) found two groups with the precipitin test.

**Symptoms.** Apparently few clinical symptoms occur in listerellosis in chickens. Paterson (1937) reported that the adult birds died suddenly, while

---

[1] Because the designation "Listerella" was found to be a homonym, Pirie (1940) proposed the new name "Listeria" for the genus. The latter is also a homonym and in order to avoid needless changes, the designation "Listerella" has been retained.— (Editor.)

a slow wasting was evident in young naturally affected chickens. It is of interest that the encephalitic symptoms which are characteristic of listerellosis in the domestic mammal have not been observed in naturally affected chickens.

Mortality in the individual flock may vary within wide limits. In the flock reported on by Seastone, sporadic cases appeared, and this was true also of the cases observed by Watkins. Paterson, however, observed heavy losses in the six flocks in which he encountered the disease. In one flock 120 out of 200 pullets died over a three-month period; in another, 191 out of 424 pullets and cockerels died; and in a third flock 8 out of 24 died. That the deaths may not have been due to Listerella alone, however, was indicated by Paterson's observation of *Salmonella pullorum* and heavy tapeworm (Davainea) infestation in the first flock, and neuro-lymphomatosis in the second. In the case reported by Hurt, Levine, and Graham (1941), ascarids, tapeworms (mostly Raillietina), coccidiosis, and iritis were present in the flock, and appeared to be more important causes of losses than Listerella. Lymphomatosis was present in the case reported by Hoffman and Lenarz. Thus there is evidence that Listerella infection in chickens may often be secondary to some other disease. Observations made so far appear to indicate that young chickens are considerably more susceptible to the disease than are adults, although these, too, may die.



FIG. 15.2. Myocardial necrosis in chickens following intravenous inoculation of Listerella. (Upper left, normal.)

**Gross pathology.** Listerella causes a septicemia in chickens. The most striking lesions which Seastone observed on autopsy of both naturally and experimentally infected chickens were massive necrosis of the heart muscle (Fig. 15.2). This was also seen by Paterson in one case and by Cole (1941). Graham, Hester, and Levine (1940a) confirmed this observation by producing both diffuse and focal necrosis of the myocardium by intravenous inoculation of chickens. According to Seastone, very little normal muscle may remain in the heart. Pericarditis is also present, and a large amount of

fluid may be present in the pericardial cavity. Microscopically, Listerella may be found within the muscle fibrils.

The lesions more commonly observed by Paterson were generalized edema, and necrotic foci in the liver. The liver lesions were apparently not present in Seastone's chickens, although he did note that the liver and spleen were congested.

The specific name *monocytogenes* was given to Listerella because of the increase in percentage of monocytes in the circulating blood of naturally affected rabbits and guinea pigs. Seastone found that a similar monocytic response is elicited in chickens.

**Diagnosis.** Listerella septicemia can be diagnosed in chickens by isolation of the causative microorganism on artificial culture media. In contrast to the domestic mammals, in which Listerella is usually confined to the central nervous system, the organism can be isolated from the abdominal organs and from the heart blood of chickens. Blood agar is probably the most desirable and convenient culture medium for this purpose.

**Therapy and prophylaxis.** No studies on treatment of avian listerellosis have been reported. However, Porter and Hale (1939) found that sulfanilamide and sulfapyridine were effective against experimental Listerella infections in mice, and it is possible that these drugs might be effective in chickens.

Graham, Morrill, and Levine (1940) were unable to immunize chickens by subcutaneous inoculation of heavy suspensions of both living and killed Listerella suspensions. Probably the disease can best be controlled by rigid sanitary measures combined with frequent culling and isolation of affected birds.

## REFERENCES

Cole, R. K.: 1941. Listeria (*Listerella*) infection in the fowl. Poultry Sci. 20:28.

Drew, R. M.: 1946. Occurrence of two immunological groups within the genus Listeria. Studies based upon precipitation reactions. Proc. Soc. Exper. Biol. and Med. 61:30.

Graham, R., Hester, H. R., and Levine, N. D.: 1940. Studies on Listerella. I. A Listerella strain isolated from a premature bovine fetus. Jour. Infect. Dis. 66:91.

———, Levine, N. D., and Morrill, C. C.: 1943. Listerellosis in domestic animals. A technical discussion of field and laboratory investigations. Univ. Ill. Agr. Exper. Sta., Bul. 499.

———, Morrill, C. C., and Levine, N. D.: 1940. Studies on Listerella. IV. Unsuccessful attempts at immunization with living and dead Listerella cultures. Cornell Vet. 30:291.

Harvey, P. C., and Faber, J. E.: 1941. Studies on the Listerella group. I. Biochemical and hemolytic reactions. Jour. Bact. 42:677.

Hoffman, H. A., and Lenarz, C.: 1942. A case of listerellosis in chickens and an additional case in sheep. Jour. Am. Vet. Med. Assn. 100:340.

Hurt, R. H., Levine, N. D., and Graham, R.: 1941. Isolation of Listeria (Listerella) from the chicken. Am. Jour. Vet. Res. 2:279.

Julianelle, L. A.: 1940. The function of *Listerella* in infection. Ann. Intern. Med. 14:608.

Lilleengen, K.: 1942. Listerellos hos tjäder. Svensk. Veter. Tidskr. 47:56, 101, 132. (Abstr. Vet. Bul. 15:4 and Bul. Inst. Past. 44:37.)

Murray, E. G. D., Webb, R. A., and Swann, M. B. R.: 1926. A disease of rabbits characterized by a large mononuclear leucocytosis, caused by a hitherto undescribed bacillus *Bacterium monocytogenes* (n. sp.). Jour. Path. and Bact. 29:407.

Pallaske, G.: 1941. Listerella-Infektion bei Hühnern in Deutschland. Berliner und Münch. tierärztl. Wochenschr. 1941:441. (Abst. Vet. Bul. 12:571.)

Paterson, J. S.: 1937. Listerella infection in fowls. Vet. Record 49:1533.

————: 1939. The present position regarding *Listerella monocytogenes* infection in animals and man. Vet. Record 51:873.

————: 1940. The antigenic structure of organisms of the genus Listerella. Jour. Path. and Bact. 51:427.

Pirie, J. H. H.: 1927. A new disease of veld rodents. "Tiger River Disease." Publ. So. African Inst. Med. Res. 3:163.

————: 1940. The genus Listerella Pirie. Science 91:383.

Porter, J. R., and Hale, W. M.: 1939. Effect of sulfanilamide and sulfapyridine on experimental infections with Listerella and Erysipelothrix in mice. Proc. Soc. Exper. Biol. and Med. 42:47.

Pothmann, E.: 1944. Listerella-Infektionen bei Schafen und Hühnern in Ostpreussen. Deut. tier. Wschr. u. Tier. Rundschau. 5250:13, 127. (Abstr. in Bul. Inst. Past. 44:38.)

Seastone, C. V.: 1935. Pathogenic organisms of the genus Listerella. Jour. Exper. Med. 62:203.

## BOTULISM

Botulism (limberneck, *Bulbar paralysis*) is a disease which may affect not only chickens and other domestic birds, but also man and the domestic mammals. It is a type of food poisoning which results from ingestion of spoiled foods in which the bacterium, *Clostridium botulinum*, has been growing and producing toxins. Other bacteria which cause food poisoning in man, such as staphylococci, have not been recognized as a cause of the same condition in domestic animals.

The earliest report of botulism as the cause of so-called limberneck in chickens was made in the United States by Dickson (1917, 1918). Spoiled canned corn, green beans, and apricots were found to have caused outbreaks in four flocks, and in three of these cases, humans also died as a result of eating the same food. Hart (1920), Wilkins and Dutcher (1920), Graham and Schwarze (1921), Doyle (1922), and Graham and Boughton (1923) also reported outbreaks of botulism in chickens. Vans Heelsbergen (1929) observed symptoms of limberneck in Holland. The disease also occurs in other species of birds. Among others, Graham and Boughton (1923) and Theiler (1927) reported it in ducks; Martinaglia (1937) in geese; Coburn and Quortrup (1938a) in turkeys; Palmer and Baker (1922), and Dobberstein and Piening (1933) in swans; and Theiler (1927) in the ostrich. It has also been described in numerous species of wild waterfowl, in which it is an important cause of death. An interesting exception reported by Kalmbach (1939) is the vulture. This bird, which lives on decaying carcasses, is tremendously resistant to *Clostridium botulinum* toxins, withstanding as much as 300,000 guinea pig minimum lethal doses. This is a tolerance to 0.04 cc. toxin injected per gram of body weight.

**Etiology.** Botulism is caused by the toxins which are a metabolic product of the growth of *Clostridium botulinum*. Some fifteen subtypes of this species have been described by Meyer and Gunnison (1929) on the basis of toxicity, agglutination, and fermentation reactions. Of these, *Clostridium botulinum* Type A and *Cl. botulinum* Type C are the most common causes of botulism in domestic birds.

*Clostridium botulinum* is an anaerobic, Gram-positive, sporeforming, large rod. It is widely dispersed in the soil, and enters food as a contaminant.

The mere presence of the organism, however, is insufficient to cause disease or to be of diagnostic significance. Only when the organism has grown, multiplied, and produced toxins does the food become dangerous. Since growth requires anaerobic conditions, canned foods furnish an excellent culture medium. Their improper sterilization at the time of canning may result in spoilage due to *Clostridium botulinum* or other organisms. It has been noted that certain foods are less liable to spoilage than others. Those which are acid, such as cherries and other acid fruits, are unfavorable for the growth of microorganisms, while canned vegetables such as corn and green beans are very favorable. Hence more cases of botulism are associated with the latter type of food.

Botulism can result from consumption of carcasses of birds which have died of the disease, and also of the maggots of the blue-bottle fly, *Lucilia caesar,* from spoiled meat. The toxin formed in the meat by Clostridium is ingested by the maggots, rendering them extremely poisonous. Wilkins and Dutcher (1920) reported the toxic character of *Lucilia* larvae, but were unable to produce limberneck in chickens with the larvae of *Calliphora vomitoria* or *Musca domestica.*

A number of carbohydrates are fermented with the production of acid and gas. Different strains and types may vary in the specific carbohydrates which they attack. In milk, Type A produces slight acidity and a slow curdling precipitate, followed by digestion and darkening. Type C produces a slowly increasing acidity, without coagulation or digestion. Type A blackens and digests meat and brain media, causing an odor of putrefaction, while Type C does not. Both types liquefy gelatin, but Type A also liquefies coagulated albumin, while Type C does not. Growth in deep liver agar medium is in the form of compact, rather lenticular discs, which may later become fluffy.

**Symptoms.** Outbreaks of botulism are usually traceable to the feeding of spoiled canned food, decomposed meat or vegetables, or spoiled grain. Feed contaminated by feces of affected birds and the bodies of birds which have died of the disease, as well as the maggots of certain flies living in such carcasses, may contain enough toxin to cause the disease.

Symptoms may appear within a few hours to a day or two after the spoiled food is eaten. The most common symptom is paralysis. The legs and wing muscles are usually the first affected. The birds become unable to walk, and the wings rest on the ground. If the neck muscles are affected the head hangs limp. This symptom is responsible for the name, "limberneck." In the early stages of botulism in poultry, the eyes are dull and partly closed. The chickens are inactive and show symptoms of weakness and unsteadiness when they move. The feathers are ruffled, and the birds refuse to eat. In mild cases the leg weakness and drowsiness may disappear, and the affected birds

recover in 2 or 3 days. In severe cases, however, death may occur in a few hours. Fatally affected birds lie in a profound coma, appearing lifeless, for several hours before death (Fig. 15.3). In the advanced stages a broken quivering of the feathers may be observed, and in some cases large numbers of feathers are shed. Looseness of the feathers is often seen in botulism. Soft, pasty feces or even diarrhea may be observed in some cases. The extent of the symptoms and the prognosis depend on the amount of toxin ingested.

**Gross pathology.** Few lesions are seen at autopsy. A slight catarrhal enteritis may be present, or small but intense inflamed hemorrhagic areas may sometimes be observed. Portions of the intestine may be dilated or distended. Other gross changes are usually lacking.

**Pathogenesis.** As stated above, all birds which have ingested *C. botulinum* toxin do not die of the intoxication. The prognosis depends primarily on the amount which has been eaten. However, this toxin is one of the most powerful



Fig. 15.3. Chicken affected with botulism.

poisons known. Bengston (1924) found that the minimum lethal dose for guinea pigs was 0.00012 mg. per kilogram when administered subcutaneously. This may be compared with the minimum lethal dose of cobra venom under similar conditions, 0.002 mg. per kg., and with that of the alkaloid aconitine, 0.06 mg. per kg.

Although it is relatively heat-stable, botulinum toxin may be destroyed by sufficient boiling. The concentration of toxin and perhaps other factors may, however, markedly affect the time necessary for its destruction. For instance, Schoenholz and Meyer (1924) found that botulinum toxin was destroyed in 6 minutes at 80° C., but that the toxin in spoiled vegetables was much more resistant, due to its high concentration and to slow heat penetration.

## WESTERN DUCK SICKNESS

For many years a disease of wild ducks and other water birds has been known to occur in the western part of the United States. It was studied by Wetmore (1918), who concluded that it was due to alkali poisoning. More

recently, however, it was shown by Kalmbach (1930) and Giltner and Couch (1930) that western duck sickness is botulism.

This disease is responsible for a tremendous number of deaths among wild waterfowl. Indeed, Kalmbach (1935a, b) considers it one of the most important causes of mortality among these birds. In 1932, it was estimated that a quarter of a million birds died at the northern end of Great Salt Lake alone. In 1910 the greatest recorded epidemic occurred in the western states, causing the death of hundreds of thousands of birds. Kalmbach (1935a) states that in recent years the epidemics have increased in frequency and that the range of the disease has also increased. Kalmbach and Gunderson (1934) list outbreaks as having occurred in Alberta and Saskatchewan in Canada; Oregon, California, Nevada, Arizona, New Mexico, Texas, Utah, Idaho, Montana, Kansas, Nebraska, North Dakota, South Dakota, and Minnesota in the United States; and Mexico. Reference should be made to their paper for a comprehensive discussion of the disease. It has since been reported in Australia by Pullar (1933, 1934) and Rose (1934), in Alberta, Canada, by Shaw and Simpson (1936), and at Tulare Lake, California, by McLean (1946). It has also been studied by Gunnison and Coleman (1932) and Coburn and Quortrup (1938b).

According to Kalmbach and Gunderson, sixty-nine species of birds belonging to twenty-one families are known to have been affected with western duck sickness. Among these are herons, geese, ducks, hawks, sandpipers, gulls, and blackbirds. Domestic birds may also die if given access to areas in which the disease is present.

Western duck sickness is caused by *Clostridium botulinum* Type C. This organism and its toxins have been isolated from the livers of affected birds, mud, decaying vegetation, and various fly larvae, and from dead fish, dead grasshoppers, and grain lying in the water. Outbreaks are associated with a number of factors, among them being prevalence of organic matter, a slight alkalinity (pH 7.5 to 9) which favors growth and toxin production, and a high temperature (37° C. optimum). The incidence of the disease is closely correlated with shallow stagnant water and mud flats. Quortrup and Holt (1941) showed that anaerobic conditions favorable for the growth of the organism are produced by decaying vegetable matter, such as aquatic plants killed by evaporation or terrestrial plants killed by flooding. They reported that potential toxin-producing areas could be detected by determining the pH and oxygen content of the water.

The possibility that *Pseudomonas aeruginosa* might be a factor in the production of botulinus toxin in these areas was suggested by Quortrup and Sudheimer (1943a). They isolated this organism frequently from the water of duck marshes and from wild duck intestines. *Ps. aeruginosa* utilizes oxygen and produces an alkaline medium, and these investigators showed that when

it is grown together with *Cl. botulinum,* an increased amount of botulinus toxin is produced.

**Diagnosis.** Diagnosis of botulism in domestic birds can be made on the basis of the paralytic symptoms, loose feathers, usual absence of marked gross lesions, and history of eating spoiled food, decaying carcasses, etc. Paralysis of the nictitating membrane of the eye occurs frequently, although it is not pathognomonic. Demonstration of the toxins in the digestive tract by feeding or inoculation of experimental animals may also be resorted to. According to Quortrup and Sudheimer (1943b) the toxin is present in such an amount in the blood of affected wild ducks that it can be detected by inoculating mice intraperitoneally with 1 ml. of serum. Mice immunized against the toxin fail to show symptoms, while nonimmunized mice may die. The organism itself can be recovered by the use of anaerobic culture media. However, it must be borne in mind that isolation of *Clostridium botulinum* from the intestinal tract does not necessarily predicate a diagnosis of botulism, since this bacterium is commonly found in soil and may be isolated from the alimentary canal of healthy, normal birds or mammals.

Among common media utilized in the cultivation of Clostridium are meat mash, brain mash, and deep tubes of liver agar. All these media should be brought to the boiling point and allowed to cool immediately before inoculation. Thioglycollate broth, which was introduced by Brewer (1940), is excellent for the cultivation of Clostridium.

**Therapy and prophylaxis.** Prevention of botulism in poultry depends primarily on feeding a wholesome ration. The feeding to chickens of spoiled canned foods, tainted meats, or decomposed vegetables should be avoided, and the danger to fowls of putrefying carcasses should be recognized.

The bodies of chickens which have died of any disease are a source of danger, and should be destroyed by burning. Since the droppings of affected birds contain toxin, sick birds should be isolated, and their droppings removed to fields that cannot be reached by poultry.

Laxatives such as castor oil or Epsom salt are of value in the treatment of exposed birds which have not yet shown symptoms of the disease. These agents can be mixed with bran in the form of a wet mash. Mildly affected chickens which cannot eat may be dosed individually with one-half ounce of castor oil. One pound of Epsom salt for each 75 to 100 chickens may be mixed in the feed for flock treatment. All other feed should be withheld, and the treatment repeated until the digestive tract is empty. The recovery of affected birds will be aided if they are kept in a cool, shady place.

Injection of antitoxin is of value in human botulism, and Quortrup and Sudheimer (1942) reported that the majority of birds affected with western duck sickness would recover if treated with Type C botulinus antitoxin. However, this measure is not generally considered practical in poultry.

## REFERENCES

Bengston, I. A.: 1924. Studies on organisms concerned as causative factors in botulism. U. S. Pub. Health Serv., Hyg. Lab. Bul. 136.

Brewer, J. H.: 1940. A clear liquid medium for the "aerobic" cultivation of anaerobes. Jour. Bact. 39:10.

Coburn, D. R., and Quortrup, E. R.: 1938a. Atypical botulism in turkeys. Jour. Am. Vet. Med. Assn. 93:385.

————: 1938b. The distribution of botulinus toxin in duck sickness areas. Trans. Third No. Am. Wildlife Conf. P. 869.

Dickson, E. C.: 1917. Botulism. A cause of limber-neck in chickens. Jour. Am. Vet. Med. Assn. 50:612.

————: 1918. Botulism. A clinical and experimental study. The Rockefeller Inst. Med. Res. Monograph No. 8.

Dobberstein, J., and Piening, C.: 1933. Beiträge zur Pathologie des Zentralnervensystems bei Tieren. I. Botulismus bei Schwänen. Berliner tierärztl. Wochnschr. 49:549.

Doyle, L. P.: 1923. Limberneck in chickens. Jour. Am. Vet. Med. Assn. 63:754.

Giltner, L. T., and Couch, J. F.: 1930. Western duck sickness and botulism. Science 72:660.

Graham, R., and Boughton, I. B.: 1923. Clostridium botulinum Type C. A pathogenic anaerobe associated with a limberneck-like disease in chickens and ducks. Illinois Agr. Exper. Sta., Bul. 246.

Graham, R., and Schwarze, H.: 1921. Avian botulism (Type A) or limber neck. Jour. Infect. Dis. 28:317.

Gunnison, J. B., and Coleman, G. E.: 1932. Clostridium botulinum Type C associated with western duck disease. Jour. Infect. Dis. 51:542.

Hart, G. H.: 1920. Clinical and case reports. Botulism in chickens. Jour. Am. Vet. Med. Assn. 57:75.

Kalmbach, E. R.: 1930. Western duck sickness produced experimentally. Science 72:658.

————: 1935a. Will botulism become a world-wide hazard to wild fowl? Jour. Am. Vet. Med. Assn. 87:183.

————: 1935b. Botulism is a factor in the decrease of western waterfowl. U. S. D. A. Yearbook. P. 140.

————: 1939. American vultures and the toxin of Clostridium botulinum. Jour. Am. Vet. Med. Assn. 94:187.

———— and Gunderson, M. F.: 1934. Western duck sickness: A form of botulism. U. S. D. A., Tech. Bul. 411.

McLean, D. D.: 1946. Duck disease at Tulare Lake. Calif. Fish and Game 32:71.

Martinaglia, G.: 1937. Some considerations regarding the health of wild animals in captivity. So. African Jour. Sci. 33:833.

Meyer, K. F. and Gunnison, J. B.: 1929. European strains of Clostridium botulinum. XXXVI. Jour. Infect. Dis. 45:96.

————: 1929. South African cultures of Clostridium botulinum and parabotulinum. XXXVII. Jour. Infect. Dis. 45:106.

————: 1929. Cultural study of an international collection of Clostridium botulinum and parabotulinum. XXXVIII. Jour. Infect. Dis. 45:119.

————: 1929. Botulism due to home canned bartlett pears. XXXIX. Jour. Infect. Dis. 45:135.

Palmer, C. C., and Baker, H. R.: 1922. Botulism (Limber Neck) in swans. Ohio State Univ., Vet. Alumni Quart. 10:93.

Piening, C.: 1933. Botulismus bei Schwänen. Tierärztl. Rundschau 39:120.

Pullar, E. M.: 1933. Limberneck (Botulism) in ducks. Australian Vet. Jour. 9:26.

————: 1934. Enzootic botulism amongst wild birds. Australian Vet. Jour. 10:128.

Quortrup, E. R., and Holt, A. L.: 1941. Detection of potential botulinus-toxin-producing areas in western duck marshes with suggestions for control. Jour. Bact. 41:363.

———— and Sudheimer, R. L.: 1942. Research notes on botulism in western marsh areas with recommendations for control. Trans. Seventh No. Am. Wildlife Conf. P. 284.

———— and Sudheimer, R. L.: 1943a. Some ecological relations of Pseudomonas aeruginosa. to Clostridium botulinum Type C. Jour. Bact. 45:551.

———— and Sudheimer, R. L.: 1943b. Detection of botulinus toxin in the blood stream of wild ducks. Jour. Am. Vet. Med. Assn. 102:264.

Rose, A. L.: 1934. Enzootic botulism amongst wild birds. Austral. Vet. Jour. 10:175.

Schoenholz, P., and Meyer, K. F.: 1924. Effect of direct sunlight, diffused daylight, and heat on potency of botulinus toxin in culture mediums and vegetable products. XXIV, Jour. Infect. Dis. 35:361.

Shaw, R. M., and Simpson, G. S.: 1936. *Clostridium botulinum* Type C in relation to duck sickness in the Province of Alberta. Jour. Bact. 32:79.

Theiler, A.: 1927. Lamsiekte (Parabotulism) in cattle in South Africa. Union So. Afr. Dept. Agr., Rep. 11–12, Dir. Vet. Ed. and Res., pt. 2:821.

van Heelsbergen, T.: 1929. Handbuch der Geflügelkrankheiten und der Geflügelzucht. Ferdinand Enke, Stuttgart.

Wetmore, A.: 1918. The duck sickness in Utah. U. S. D. A., Bul. 672.

Wilkins, S. D., and Dutcher, R. A.: 1920. Limberneck in poultry. Jour. Am. Vet. Med. Assn. 57:653.

# ERYSIPELOTHRIX SEPTICEMIA
## (Geflügelrotlauf)

The occurrence of a septicemia associated with *Erysipelothrix rhusiopathiae*, the causative agent of swine erysipelas, has been reported in many species of birds. Jarosch (1905) was the first to recognize this disease in birds, isolating the organism from a turkey. Subsequently the disease has been found to occur also in the chicken, duck, pigeon, pheasant, quail, peacock, and birds in zoological gardens. Since Jarosch's initial paper, erysipelas has been described in turkeys by Eber (1921), Beaudette and Hudson (1936), Madsen (1937), Hoffman and Hinshaw (1938), Van Roekel, Bullis, and Clarke (1938), Rosenwald and Dickinson (1939, 1941), Rosenwald (1940), Schlotthauer and Thompson (1940), and Lindenmayer and Hamilton (1942). According to Grey (1947b), there have been thirty-nine recorded outbreaks in turkeys in twelve states: Colorado, Connecticut, Iowa, Massachusetts, Minnesota, Nebraska, New Jersey, New York, Oregon, Utah, Virginia, and Washington. The largest number, sixteen, was recorded from Oregon. In addition, Moore (1947) stated that eight outbreaks in turkeys were diagnosed in New York in the single year, 1946. The disease was first described from the chicken by Hausser (1909), and has since been reported in this bird by Schipp (1910), Broll (1911), Pfaff (1921), Reinhardt (1924), Scholl and Jacquart (1926), van Heelsbergen (1929), Schmidt-Hoensdorf (1931), Sparapani (1938), and Breed (1943). Poels (1919) first encountered erysipelas septicemia in ducks, and this finding was confirmed by Eber (1921), Scholl and Jacquart (1926), Werner (1932), Horstmann (1938), Graham, Levine, and Hester (1939), White and Henley (1942), and Doria (1943). The disease was recognized in the pigeon by Poels (1919), in the quail by Jármai (1920) and Waller (1939), in the pheasant by Vianello (1938), Morgan (according to Waller, 1939), and Szabó (1943), in the peacock by Greener (1939), and in birds in zoological gardens by Jármai (1920) and Schmidt-Hoensdorf (1931). De la Villa (1934) reported that the green finch was susceptible to Erysipelothrix infection, while Urbain, Nouvel, and Roth (1943) isolated the organism from a ring-necked parakeet

(*Palaeornis torquata*). The same organism, or a very similar one, has also been found in the sheep, ox, horse, dog, guinea pig, mouse, mink, and several species of fish (Van Es and McGrath, 1936; Grey, 1947a).

**Etiology.** The etiologic agent, *Erysipelothrix rhusiopathiae*, can be isolated culturally from many of the organs of affected birds. Since the disease is a septicemia, the heart blood, liver, pericardial fluid, and pathologic accumulations of fluid usually contain large numbers of the organisms. The bacterium has also been isolated from diphtheritic membranes on the pharyngeal and nasal mucosae. While isolation may be accomplished on plain nutrient agar, it is probably more uniformly successful if blood or serum is added to the medium.



*E. rhusiopathiae* on first isolation is a Gram-positive rod, but after continued transfer on artificial culture media it may become Gram-negative. In the animal body and when first isolated the organisms may be relatively short (1–2μ long), but on cultivation they ordinarily are in the form of slender filaments 4–15μ long (Fig. 15.4). Branching of the filaments has been described. The organism forms no spores, and is non-motile. The colonies are round and

FIG. 15.4. *Erysipelothrix rhusiopathiae*, agar culture. ×2,000. (From Nowak: Documenta Microbiologica, courtesy Gustav Fischer.)

translucent, usually about 1 mm. in diameter. On first isolation they are smooth, but on continued transfer larger, rough colonies may appear. Gelatin is not liquefied, but a typical "test tube brush" growth occurs along the line of the stab. Litmus milk may be slightly acidified. A narrow zone of green hemolysis forms on blood agar. Neither indol nor acetyl methyl carbinol are formed, but hydrogen sulfide is produced. Gas is not formed from carbohydrate media. According to Karlson (1938), acid is formed in dextrose, lactose, galactose, and levulose, while mannose and cellobiose are fermented late, and no acid is formed from arabinose, xylose, rhamnose, maltose, melibiose, sucrose, trehalose, raffinose, melezitose, dextrin, starch, inulin, amygdalin, salicin, glycerol, erythritol, adonitol, mannitol, sorbitol, dulcitol, or inositol. Maximum growth occurs at a pH of 7.6 at 37.5° C.

**Symptoms and pathology.** *Turkey.* Erysipelothrix septicemia usually affects poults from four to seven months of age, although Rosenwald (1940) reported it in a 7-day-old poult. It usually appears in the fall, most outbreaks which have been reported having occurred in October and November. Losses may be fairly heavy, mortality of from 2½ to 25 per cent having been

reported in different flocks. An interesting characteristic of this disease is the fact that male birds are apparently much more susceptible than females. This differential sex incidence has been reported by a number of writers. Madsen (1937), for instance, reported that out of 325 turkeys which died in a flock of 1,200, only 25 were females, while Rosenwald and Dickinson (1941) reported that 82.1 per cent of the turkeys affected in outbreaks in sixteen flocks were males.

Symptoms include general weakness and listlessness, inappetence, and sometimes a yellowish or greenish diarrhea. Affected birds stand or crouch with lowered head, drooping wings and tail, and ruffled feathers. The skin and wattles may be cyanotic. Dyspnea and an ulcerated nasal mucosa may be present.

At autopsy, the most characteristic lesions are petechial and diffuse hemorrhages in many of the tissues and organs. They have been reported in the abdominal, pectoral, and femoral muscles, fascia, peritracheal tissues, pleura, peritoneum, pericardium, heart, lungs, spleen, and small intestine. Rosenwald and Dickinson (1941) consider the most pathognomonic lesion to be a turgid, reddish-purple caruncle. The liver is enlarged, congested, and often friable or mottled. In some cases necrotic foci may be present in this organ. The spleen is usually enlarged, congested, and friable, and hemorrhages or necrotic foci may be observed. The kidneys may be enlarged and congested. The lungs may be congested or brownish in color. A catarrhal exudate which may be sanguineous is present in the intestine, and the intestinal mucosa may be inflamed, edematous, hemorrhagic, or even necrotic. The mesenteric and other blood vessels are often engorged. A nasopharyngeal catarrh has been observed in some cases. Inflammation of the mucosa of the proventriculus has been reported, and a swollen joint was noted in one case by Beaudette and Hudson (1936). On the other hand, in the cases in week-old poults reported by Rosenwald (1940), characteristic lesions were absent on autopsy.

*Chicken.* Symptoms of lassitude, inappetence, and diarrhea may be present in the chicken. The pathological-anatomical changes induced in this bird by *Erysipelothrix rhusiopathiae* are usually not so marked as in the turkey. In the epidemic reported by Schipp (1910), only an enteritis and a parenchymatous degeneration of the heart muscle were found. Broll (1911) reported a superficial streaky hemorrhage under the epicardium, cloudy swelling of the parenchyma and hemorrhagic enteritis. Pfaff (1921) found extensive changes in a seven-month-old cockerel which had been sick about 8 days. At autopsy, a patchy fibrinopurulent exudate was found on the pleura, and necrotic foci the size of a hemp seed were present in the edematous lung. The liver was degenerated, and the spleen enlarged. The esophageal and tracheal mucosae were swollen and roughened. The proven-

triculus was adherent to the left wall of the thoracic cavity; the gizzard wall was thickened and friable, and its mucosa showed necrosis and ulceration in the thickened area. The mucosa of the small intestine exhibited catarrhal edema, and the posterior portion was heavily injected and covered with petechiae. The ceca were enlarged, with edematous mucosa containing numerous petechiae and small yellowish nodules visible through the serosa. The cloacal mucosa was markedly hyperemic.

In other affected chickens the lesions were not so pronounced. Autopsy showed only swelling of the proventricular and intestinal mucosae, a fibrin-opurulent exudate in the cecum, or small hemp-seed size necrotic foci in the liver. The intestinal mucosa was injected, and necrotic nodules were often present in the cecum. In very young birds there were seen only catarrhal enteritis and enlargement of the spleen.

Of interest is the report by Sparapani (1938) of a paralysis of chickens due to lumbar meningitis caused by *Erysipelothrix rhusiopathiae.*

*Duck.* Ducklings two months or more of age are most susceptible to Ery-sipelothrix septicemia. Losses may be quite high. In one flock of 46,000, Graham, Levine, and Hester (1939) reported a loss of almost 25 per cent. In the duck the gross pathologic lesions are similar to those in the turkey and chicken. A serofibrinous exudate may be present in the air sacs, and the lungs are often congested. The liver is often enlarged, friable, mottled, and may contain numerous yellowish pin-point foci. The spleen is usually congested, enlarged, and soft. Petechial hemorrhages may be present in the heart. Areas of congestion are often found in the intestine and catarrhal enteritis may be observed. Dark, congested areas in the webs of the feet, and chronic enlargement of the femorotibial articulations have also been reported. Congestion may be found in many of the organs, and petechiation of the muscles may also occur.

The symptoms and lesions caused by *Erysipelothrix rhusiopathiae* in other species of birds are similar to those described above.

**Pathogenesis.** The pathogenicity of *Erysipelothrix rhusiopathiae* for pigeons is well established. Indeed, in some laboratories, inoculation of these birds is a routine procedure for the diagnosis of swine erysipelas. However, the ability of this organism to cause disease in other birds in the absence of contributory factors has been questioned. In the flock of turkeys reported on by Beaudette and Hudson (1936), the existence of blackhead had been suspected by the owner, and all the birds were given iodine per os. Heavy losses started the next day, and these writers consider that the infection may have been spread through the flock by the contaminated catheter. Among predisposing factors they mention confinement and overcrowding. Other writers have emphasized damp or inclement weather, drafts, sudden temperature changes, and lack of sanitation as contributory agents. Other diseases,

malnutrition, in fact any condition which lowers the vitality of the birds may increase their susceptibility to this disease. Of interest in this connection is the report of Marinelli (1928) that the feeding of polished rice lowers the resistance of pigeons to Erysipelothrix. It is noteworthy that most of the outbreaks of Erysipelothrix septicemia reported have occurred in the fall and winter months.

While the pathogenicity of *E. rhusiopathiae* is about the same for the duck as for the turkey, it is definitely lower for the chicken. Most of the cases in this species have been sporadic losses rather than epidemic in nature. In the outbreak reported by Breed (1943), two-thirds of a flock of 300 six-month-old chickens died, but both *E. rhusiopathiae* and *Pasteurella avicida* were isolated from affected birds. Experimental inoculations of the organism by a number of workers give further confirmation of the relatively greater resistance to infection of the chicken. Van Heelsbergen (1929) observed three cases of fowl Erysipelothrix septicemia in the ten years before 1929. Two birds had suffered from tuberculosis, and the third had died of leukemia.

**Diagnosis.** While extensive petechiation is quite characteristic of Erysipelothrix septicemia in birds, reliance cannot be placed upon this lesion alone in arriving at a diagnosis. A positive diagnosis can be made only by isolating the causative microorganism from the tissues of affected birds by cultural methods, and identifying it. For best results, cultures of heart blood, liver, or other organs should be seeded on blood or serum agar. It is recommended that the agglutination test be used for positive identification of the organism recovered.

A presumptive diagnosis can often be made by examination of thin blood smears stained by Gram's method. The organisms may be abundant in the heart blood or spleen. Filamentous forms are usually absent in the animal tissues.

A serological test might be helpful, since it has been reported by a number of workers that high agglutinin titers may result from natural or experimental infection.

**Therapy.** The sulfonamides are apparently valueless for treatment of *E. rhusiopathiae* infections. Sulfanilamide, sulfapyridine, sulfathiazole, irgafen ($N_1$-3-4-dimethyl benzoyl sulfanilamide), sulfaguanidine, phthalyl-sulfathiazole, 3-sulfanilamidobenzamide and sulfetrone have all been found ineffective when tested on infected mice (Konst, 1945; Frei and Jezierski, 1945; Woodbine, 1946).

Penicillin has been found to be active against *E. rhusiopathiae*, repeated injections being most effective. Both Heilman and Herrell (1944) and Grey (1947a) found that 0.1 Oxford unit per ml. prevented growth of the organism in culture, and that this antibiotic was effective in the treatment of ex-

perimental infections in mice. On the other hand Woodbine (1946) and Woodbine with Cheeseman (1947) reported that extremely high doses of penicillin were necessary to protect mice. Van Es, Olney, and Blore (1945) found penicillin effective when given in repeated injections in the treatment of pigeons infected with *E. rhusiopathiae*. Grey (1947b) reported that the mortality among infected turkeys was reduced 90 per cent by injection into the wattles on 4 consecutive days of doses of 20,000 units of penicillin in peanut oil.

Streptomycin is not as effective as penicillin. Grey (1947c) found that 140,000 mcg. in distilled water injected into the wattle in a single dose was necessary to prevent death in artificially exposed turkeys, while Woodbine with Cheeseman (1947) found that streptomycin was about one-fifth as effective as penicillin in mice and one-hundredth as effective in vitro.

Treatment with antiserum may be of some value, but only if it is administered very early or before clinical symptoms appear. Sparapani (1938) reported the successful use of swine erysipelas antiserum in the outbreak in chickens which he studied. White and Henley (1942) reported that death was prevented in ducks following early treatment with two doses of antiserum administered intramuscularly on consecutive days. Breed (1943) stated that losses in the flock of chickens he studied ceased following administration of 2 ml. doses of antiserum, but that birds already sick were not helped. Grey (1947b) found that a single 10 ml. injection of swine erysipelas antiserum reduced mortality from the infection if it was administered sufficiently early. Lindenmayer and Hamilton (1942) considered that the use of formolized serum from an affected turkey was helpful in treatment of other turkeys, but their experiment was uncontrolled. On the other hand, attempts by Graham, Levine, and Hester (1939) to protect ducklings against the disease on contaminated premises by the use of antisera and bacterins were unsuccessful. Rosenwald and Dickinson (1941) reported that the use of commercial anti-swine erysipelas serum was impractical in the treatment or prevention of the disease in turkeys. Furthermore, varying results have been obtained by different workers on the use of antisera for protection against artificial exposure. In most of these studies mice or pigeons were used as the experimental animals.

## REFERENCES

Beaudette, F. R., and Hudson, C. B.: 1936. An outbreak of acute swine erysipelas infection in turkeys. Jour. Am. Vet. Med. Assn. 88:475.

Breed, F.: 1943. *Erysipelothrix rhusiopathiae* and *Pasteurella avicida* in chickens. Vet. Med. 38:430.

Broll, R.: 1911. Über das Vorkommen von rotlaufähnlichen Bakterien beim Rinde und Huhne. Berliner tierärztl. Wochnschr. 27:41.

Doria, C.: 1943. Su un'enzoozia di mal rossino nelle anitre. Nuova Vet. 21:72. (Abstr. Vet. Bul. 16:39.)

Eber, A.: 1921. Geflügel-Rotlauf (Rotlauf-Septikämie der Vögel). Deutsch. tierärztl. Wochenschr. 29:295.

Frei, W., and Jezierski, A.: 1945. Chemothreapeutische Versuche mit Sulfanilamiden bei der Geflügelcholera- und Rotlaufinfektion der weissen Maus. Schweiz. Arch. Tierheilk. 87:136. (Abstr. Vet. Bul. 17:204.)

Graham, R., Levine, N. D., and Hester, H. R.: 1939. *Erysipelothrix rhusiopathiae* associated with a fatal disease in ducks. Jour. Am. Vet. Med. Assn. 95:211.

Greener, A. W.: 1939. Infection of a peacock with *Erysipelothrix rhusiopathiae*, followed by a case of human erysipeloid. Brit. Jour. Dermat. 51:372.

Grey, C. G.: 1947a. Effects of penicillin on *Erysipelothrix rhusiopathiae* and on mice infected with that organism. Vet. Med. 42:74.

——: 1947b. Penicillin in the treatment of *Erysipelothrix rhusiopathiae*-infected turkeys. Vet. Med. 42:177.

——: 1947c. Streptomycin in the treatment of *Erysipelothrix rhusiopathiae*-infected turkeys. Vet. Med. 42:216.

Hausser, A.: 1909. Bakteriologische Untersuchungen über Geflügeldiphtherie. Zentralbl. f. Bakt., I. Orig. 48: 535.

Heilman, F. R., and Herrell, W. E.: 1944. Penicillin in the treatment of experimental infections due to *Erysipelothrix rhusiopathiae*. Proc. Mayo Clin. 19:340.

Hoffman, H. A., and Hinshaw, W. R.: 1938. Erysipelas of turkeys. Poultry Sci. 18:443.

Horstmann, H.: 1938. Ein Beitrag zum Rotlauf bei Enten. Zeitschr. f. Infekt-Krankh. der Haustiere. 53:106.

Jármai, K.: 1920. Das Vorkommen der Rotlaufbazillen bei Vögeln. Allotorvosi Lapok. 1919. 8:57 (Abstr. Berliner tierärztl. Wochnschr. 36:17) .

Jarosch, L. W.: 1905. Ueber Septikämie der Truthühner. Oesterr. Monatschr. Tierheilk. 30:197.

Karlson, A. G.: 1938. The cultural characteristics of *Erysipelothrix rhusiopathiae*. Jour. Bact. 35:205.

Konst, H.: 1945. Chemotherapy of swine erysipelas. Trials using sulfanilamide, sulfapyridine, and sulfathiazole in experimental infection of mice. Canad. Jour. Comp. Med. 9:135.

Lindenmayer, J. E., and Hamilton, C. M.: 1942. Treatment of swine erysipelas in turkeys with serum from a turkey infected with *Erysipelothrix rhusiopathiae*. Jour. Am. Vet. Med. Assn. 100:212.

Madsen, D. E.: 1937. An erysipelas outbreak in turkeys. Jour. Am. Vet. Med. Assn. 91:206.

Marinelli, G.: 1928. Le infezioni da mal rossino e da colera dei polli nei colombi tenuti a riso brillato. Folia Med. 14:1478.

Meyer, A.: 1928. Vergleichende Untersuchungen über Schweine- und Geflügelrotlaufbakterien. Deutsch. tierärztl. Wochnschr. 36:334.

Moore, E. N.: 1947. Diseases of turkeys in New York. Cornell Vet. 37:112.

Pfaff, F.: 1921. Schweinerotlaufbakterien als Erreger einer chronischen Hühnerseuche. Zeitschr. f. Infekt-Krankh. der Haustiere 22:293.

Poels, J.: 1919. Rotlauf bei Tauben und Enten und Stammunterschiede bei Rotlaufbazillen. Folia Microbiol. 5:1.

Reinhardt, R.: 1924. Septikämische Erkrankungen bei Schafen, verursacht durch Schweinerotlaufbazillen (including poultry) . Monatshefte für prakt. Tierheilk. 34:155.

Rosenwald, A. S.: 1940. ·Swine erysipelas in a week-old turkey poult. Jour. Am. Vet. Med. Assn. 96:268.

—— and Dickinson, E. M.: 1939. A report of swine erysipelas in turkeys. Cornell Vet. 29:51.

—— and Dickinson, E. M.: 1941. Swine erysipelas in turkeys. Am. Jour. Vet. Res. 2:202.

Schipp, C.: 1910. Zur Biologie des Schweinerotlaufbazillus und zweier morphologisch gleicher Septikämieerreger. Deutsch. tierärztl. Wochnschr. 18:97.

Schlotthauer, C. F., and Thompson, L.: 1940. The occurrence of erysipelas in turkeys. Jour. Am. Vet. Med. Assn. 96:103.

Schmidt-Hoensdorf, F.: 1931. Rotlauferkrankungen bei Vogeln im Anschluss an Schweinerotlauf und Mäuseseptikämie. Deutsch. tierärztl. Wochnschr. 39:196.

Scholl, M. A., and Jacquart, M.: 1926. Enzootie chez la poule et le canard, provoquée par le bacille du rouget. L'écho vétérinaire 55:49.

Sparapani, G.: 1938. Casi interessanti di meningite lombare in polli Sussex millefiore causata dal bacillo del mal rossino dei suini. Coltura Avicola 1938:139, 158. (Abstr. Int. Rev. Poult. Sci. 11:238.)

Szabó, B.: 1943. *Erysipelothrix rhusiopathiae* infection in pheasants (trans. title) . Allatorv. Lapok. 17:100. (Abstr. Vet. Bul. 16:387.)

Urbain, A., Nouvel, J., and Roth, P.: 1943. Septicémie à bacille du rouget chez une perruche (*Palaeornis torquata*, L.) . Bul. Acad. Vét. France 16 (n.s.) :136. (Abstr. Vet. Bul. 15:72.)

Van Es, L., and McGrath, C. B.: 1936. Swine Erysipelas. Nebr. Agr. Exper. Sta., Res. Bul. 84.

——, Olney, J. F., and Blore, I. C.: 1945. The effects of penicillin on *E. rhusiopathiae*-infected pigeons. Nebr. Agr. Exper. Sta., Res. Bul. 141.

van Heelsbergen, T.: 1929. Handbuch der Geflügelkrankheiten und der Geflügelzucht. Ferdinand Enke, Stuttgart.

Van Roekel, H., Bullis, K. L., and Clarke, M. K.: 1938. Erysipelas outbreaks in turkey flocks. Jour. Am. Vet. Med. Assn. 92:403.

Vianello, G.: 1938. Un'enzoozia da mal rossino nei fagiani. La Clinica Véterinara. 61:234.

de la Villa, G. C.: 1934. Nota sobre la sensibilidád del pájaro al bacilo *Erysipelothrix rhusiopathiae*. Trab. Inst. Biol. Anim. Madrid 2:330. (Abstr. Vet. Bul. 5:789.)

Waller, E. F.: 1939. Erysipelothrix infection in a quail. Jour. Am. Vet. Med. Assn. 95:512.

Werner, F.: 1932. Ein Beitrag zum Geflügelrotlauf bei Enten. Deutsch. tierärztl. Wochnschr. 40:148.

White, E. G., and Henley, F. A.: 1942. *Erysipelothrix rhusiopathiae* associated with disease in ducks. Vet. Rec. 54:127.

Woodbine, M.: 1946. Chemotherapy of *Erysipelothrix rhusiopathiae* infections in mice. Vet. Jour. 102:88.

——— with asst. of Cheeseman, M. W.: 1947. Chemotherapy of *Erysipelothrix rhusiopathiae* infections in mice with streptomycin. Vet. Jour. 103:149.

## GOOSE INFLUENZA

### (*Septicaemia anserum exsudativa*)

Goose influenza was first described in some detail by Riemer (1904) as the cause of a fatal disease of geese in Germany, and a specific bacterium was isolated from the affected birds. A similar microorganism was found in geese by Bugge (1908). This disease was later encountered in several German provinces by a number of investigators. Their data indicate that the disease was probably introduced into Germany by Russian and Polish geese. The disease of geese described by M'Fadyean (1902) in England was probably goose influenza. The hemorrhagic septicemia observed by Fiorentini (1896) in geese and swans was, however, due to a different organism, a bipolar staining, motile coliform rod, pathogenic for chickens, pigeons, rabbits, guinea pigs, geese, and ducks. The influenza bacterium, on the other hand, is pathogenic only for geese. Although this disease has not been reported in North America, the writers (unpublished data) isolated an organism from a young goose in Illinois which was similar in preliminary tests to the causative agent of goose influenza.

**Etiology.** The organism causing goose influenza appears in the animal body as a small rod, frequently resembling a diplococcus. It varies in size from 0.5 to 1.5 by 0.5µ. It can always be found in large numbers in the heart blood, pericardial fluid, and fibrinous exudates. It is Gram-negative, forms no spores, and is nonmotile. While it can be rather easily isolated from an affected bird on plain nutrient agar, a medium containing hemoglobin is necessary for further transfers. Once adapted to artificial culture media, however, it will grow on plain agar.

Small, white, circular colonies are formed on gelatin plates, while in gelatin stabs a slight infundibuliform liquefaction occurs which becomes complete in several weeks. On nutrient agar, circular, transparent, smooth, homogeneous slightly viscid colonies are formed. They are grayish white (bluish by transmitted light) at first, but become brownish-yellow with age. In broth a uniform turbidity is produced, and a slight sediment is formed after a few days. A pellicle may or may not be formed. Indol is usually not

formed. Litmus milk is unchanged. Slight acid may be produced from dextrose, but not from lactose. Hydrogen sulfide is formed. On coagulated blood serum, a yellowish-white growth appears, and the medium later becomes brownish and still later is liquefied. No growth occurs on Drigalski-Conradi or Endo media. The optimum temperature is about 37.5° C. No growth occurs below 15°, while an exposure to 56° C. for 5 minutes kills the organisms. Cultures are viable only a relatively short time.

The systematic position of this organism is still uncertain. Riemer (1904) named it *Bacillus septicaemiae anserum exsudativae,* while Bergey *et al.* (1939) placed it in the genus Shigella, giving it the name *Shigella septicaemiae.* According to Frosch and Bierbaum (1909), Löffler (1910), and Schlüter (1936), however, it belongs in the Hemophilus group; this latter position appears to be the correct one.

**Symptoms.** Both young and old geese are susceptible to the disease, while ducks, chickens, and turkeys are not affected. The disease generally appears in May and the first half of June, sometimes reappears in the latter part of August and September. At the beginning of an outbreak, a few cases and some deaths are observed, but later a high percentage of the flock may become affected. At first the disease is found in the young geese, but as the epidemic progresses older birds are also affected. The mortality in young birds is especially high, often 70 to 90 per cent. The disease usually disappears from the flock in two to four weeks.

Few affected birds survive, and these generally exhibit paralysis or leg weakness for some time.

The first striking symptom is a decrease in appetite. The birds gradually become weaker, squat with ruffled feathers, and keep apart from the rest of the flock. According to Bugge, diarrhea sets in 12–24 hours before death. The birds strike with their legs and head, stagger while walking, and breathe rapidly. The beak is opened wide, and a snoring noise is emitted. Death may come either gradually or quite suddenly. The course of the disease is ordinarily 2 to 5 days, although sometimes the birds may die within an hour after symptoms appear.

**Pathology.** In this disease two post-mortem pictures have been described which are in partial agreement. Riemer described an exudative septicemia, while Bugge emphasized the inflammation of the air sacs and the lung lesions. Both Riemer (1904) and Eber (1921) found at autopsy only a tender, lightly adhering film on the liver surface, fine fibrinous threads on the epicardium, and a small amount of a turbid, serous exudate in the pericardium. No other morbid changes in the organs or tissues were demonstrated. According to Bugge, the roughened surface of the air sacs is covered by a yellowish film which can be easily pulled off, and the inner surfaces are covered by thick, coherent, whitish-yellow fibrin masses frequently distributed in reticular

fashion. Since these masses may continue on to the lateral and posterior borders of the lung, numerous yellow nodules from the size of a pinhead to that of a pea are visible through the healthy rosy-red lung tissue at these places. The peripheral bronchi are filled with yellowish fibrinous masses. In a few cases hemorrhages in the intestinal mucosa and enlargement of the spleen, liver and kidneys are observed.

**Pathogenicity.** The organism of goose influenza is pathogenic for geese, but rabbits, gray and white rats, mice, ducks, chickens, and pigeons are not affected by it. Guinea pigs may be killed by a large inoculum of the bacteria. Cultures lose their virulence rather rapidly if they are not passed through geese regularly.

**Diagnosis.** In dead geese the disease is recognized rather easily by the characteristic fibrinous inflammation of the serous membranes or of the air sacs as well as by symptoms of a general septicemia. The diagnosis is confirmed by recognition of the bacteria in stained smears from affected areas or heart blood, or by isolation and identification of the causative microorganism.

**Therapy.** While vaccination with an autogenous bacterin may be attempted, the main emphasis in the control of goose influenza must be placed on the application of hygienic measures. Newly acquired birds should be held under quarantine. Sick birds should be separated from the rest of the flock or slaughtered, and all carcasses should be destroyed by burning. Houses and runs should be disinfected.

## REFERENCES

Bergey, et al.: 1939. Manual of Determinative Bacteriology. 5th Edition. Williams and Wilkins, Baltimore.

Bugge, G.: 1908. Ansteckende Luftsackentzündung der Gänse. Zeitschr. f. Infekt-Krankh. d. Haustiere 3:470.

Eber, A.: 1921. Gänse-Influenza (exsudative Septikämie und ansteckende Luftsackentzündung der Gänse). Deutsch. tierärztl. Wochnschr. 29:187.

Fiorentini, A.: 1896. Hämorrhagische Septikämie der Schwäne. Zentralbl. f. Bakt. etc. I. Orig. 19:932.

Frosch, P., and Bierbaum, K.: 1909. Ueber eine durch den Bacillus septicaemia anserum exsudativae (Riemer) bedingte Gänseseuche. Zentralbl. f. Bakt. etc. I. Orig. 52:433.

Löffler, F.: 1910. Ueber eine in Jahre 1904 in Klein-Kiesow bei Greifswald beobachtete Gänseseuche. Arch. f. wiss. u. prakt. Tierheilk. 36:289. Suppl. Bd.

M'Fadyean, J.: 1902. A remarkable outbreak of goose septicaemia. Jour. Comp. Path. Therap. 15:162.

Miessner, H. and Berge, R.: 1923. Septicaemia anserum exsudativa (Gänseinfluenza). Deutsch. tierärztl. Wochnschr. 31:539.

Pröscholdt: 1919. Gänseinfluenza. Berliner tierärztl. Wochnschr. 35:261.

Reinhardt, R.: 1921. Untersuchungen über die Septicaemia anserum exsudativa. Zeitschr. f. Infekt-Krankh. d. Haustiere 21:257.

Riemer: 1904. Kurze Mitteilung über eine bei Gänsen beobachtete exsudative Septikämie und deren Erreger. Zentralbl. f. Bakt. etc. I. Orig. 37:641.

Schlüter, W.: 1936. Beitrag zur serologischen Differenzierung hämoglobinophiler Bakterien. Zentralbl. f. Bakt. etc. I. Orig. 136:362.

CHAPTER SIXTEEN

# STREPTOCOCCOSIS, STAPHYLOCOCCOSIS, ARTHRITIS COLIBACILLOSIS, AND HJÄRRE'S DISEASE

*By J. L. Noordsy, Veterinary Research Institute,*
*Iowa State College, Ames, Iowa*

## AVIAN STREPTOCOCCOSIS

✻   ✻   ✻

The importance of streptococcic infections in birds is far greater than is commonly attributed to this pathogenic condition. The acute septicemic form responsible for considerable losses has been commonly referred to as "apoplectiform septicemia" (Nörgaard and Mohler, 1902). The more chronic form of the disease is sometimes designated as "sleeping sickness of chickens" (Dammann and Manegold, 1905). Avian streptococcosis is also known as "streptomycosis of fowls" (Hutyra and Marek, 1938).

**History.** The first report of streptococcosis in chickens recorded in North America was that of Nörgaard and Mohler (1902). This outbreak occurred on a farm in Loudoun County, Virginia, and proved to be an acute septicemic form which resulted in heavy losses. Death occurred suddenly without premonitory symptoms, resulting in a mortality of 100 per cent of the affected birds and 92 per cent of the entire flock. This disease was designated as "apoplectiform septicemia of chickens."

The report of Dammann and Manegold (1905) records the outbreak of a more chronic form of this disease in Germany. The organism responsible for this infection was found to be a capsulated streptococcus which was at that time designated as *Streptococcus capsulatus gallinarum.* Only eight of 100 chickens were lost, and the outbreak lasted over a period of three months. Experimental studies indicated that pigeons, white mice, rabbits, and lambs were susceptible to infection while dogs, ducks, and guinea pigs were not. Birds infected under experimental conditions lived from 7 to 50 days following inoculation. The characteristic sleepiness manifested by the diseased birds suggested the term "sleeping sickness."

Another outbreak of a similar infection of fowl in Germany was reported by Greve (1908). This apparently was an acute form of the disease terminating fatally in 12 to 24 hours following the initial symptoms. The etiologic agent was found to be a streptococcus which was believed to be *Streptococcus*

*capsulatus gallinarum* described by Dammann and Manegold (1905). The mortality was much higher in the young birds than in mature fowl.

A highly acute outbreak of streptococcosis was reported by Magnusson (1910) in Sweden. The infection spread rapidly throughout the flock with a mortality of about 65 per cent within a period of 14 days. The infection spread to neighboring farms with a resulting mortality of over 30 per cent. A Gram-positive, short-chained streptococcus which was assumed to be *Streptococcus capsulatus gallinarum* described by Dammann and Manegold (1905) was isolated from infected birds. The mortality was much higher among the young birds.

Experimental investigations indicated that chickens, mice, pigeons, and rabbits were highly susceptible while guinea pigs and young swine were resistant to infection. A much greater susceptibility was observed in Leghorn than in Plymouth Rock chickens. A single case of joint abscess with suppuration in an experimental dog was reported which was the only instance of this organism developing pyogenic characteristics. Bacteriological studies made by Magnusson resulted in the isolation of two forms or strains which apparently possessed some specificity for certain species. The formation of capsules on these organisms, as reported by Dammann and Manegold (1905), could not be confirmed. The apoplectiform symptoms and subdural exudate described by Nörgaard and Mohler (1902) were not observed. Magnusson designated the condition "streptomycosis of hens." Another highly acute streptococcic septicemia of fowl in Sweden was reported by Bergman (1907). The etiologic agent was isolated from the infected birds, and bacteriologic investigations were made of the cultures. The streptococci isolated were reported as Gram-negative, whereas some previous investigators found the organism to be Gram-positive.

A report on outbreaks of chronic streptococcic peritonitis of hens occurring in Minnesota was published by Kernkamp (1927). The condition developed on two farms 125 miles apart. Pure cultures of streptococci were isolated from tissues of infected birds. These organisms did not show the presence of either flagella or capsules and were only slightly Gram-positive. The reactions of this type of streptococcus in the carbohydrates and on blood agar classified it in the *Streptococcus pyogenes* (Rosenbach) group. The most prominent pathological lesion was a chronic peritonitis accompanied by considerable quantities of fibrinous exudate in the peritoneal cavity.

Acute streptococcic septicemia of fowl was reported in New Jersey by Hudson (1933). A high mortality occurred among infected birds. Some of the pathologic lesions found included hemorrhagic discoloration of the breast muscles, reddish fluid exudate in the peritoneal cavity, hemorrhagic enteritis, lung congestion, as well as enlargement of the liver and spleen. Bacteriologic

examination of the cultures grown on blood agar revealed short-chained streptococci. The organism was reported to be Gram-positive and non-capsule forming. Subsequently, Edwards (1934), working with Hudson's strain of streptococci, definitely observed capsule formation. Birds inoculated under experimental conditions with cultures of these organisms developed symptoms similar to those described by Nörgaard and Mohler (1902).

A chronic infection of hens due to a hemolytic streptococcus was observed by Edwards and Hull (1937). The disease was reproduced under experimental conditions causing a wide variety of symptoms and pathologic changes during its course. The only constant pathological changes reported were exudative peritonitis and salpingitis accompanied by the formation of concretions in the oviduct. The organisms isolated from infected birds were characteristic of the *Streptococcus pyogenes* group. Edwards and Hull (1929) isolated a streptococcus from abnormal ova of hens believed to have been infected with *Salmonella pullorum*. Beaudette and Hudson (1931) reported additional cases of streptococcic infections in fowl. Gray (1939) isolated streptococci together with other organisms from the nerves of fowl which developed symptoms of "fowl paralysis." The report of Genest and Nadeau (1944) described an outbreak of acute streptococcus infection which occurred in Quebec. Gwatkin (1946), referring to Genest and Nadeau's report, suggested the possibility of virus infection and expressed some doubt as to pathogenic action created by the streptococci which were isolated from infected fowl. Losses among young geese and ducklings caused by an acute form of streptococcosis were observed by Hodosy (1944). Two distinct diseases were described by Ward and Gallagher (1920) termed "apoplectiform septicemia" and "sleeping sickness" which were undoubtedly applied to the acute and chronic forms of streptococcosis, respectively.

**Etiology.** The etiologic agent responsible for this disease is designated as *Streptococcus gallinarum*. Because of insufficient scientific data, this organism is not included in the classification of "animal pyogenes" suggested by Sherman (1937). There seems to be considerable difference of opinion, especially among the early investigators, regarding the fermentation of sorbitol and the presence of capsules. Edwards (1933) states that 96 per cent of the animal streptococci studied thus far are sorbitol-fermenters and produce capsules. The organism is described by Merchant (1946) as Gram-positive, usually composed of six to eight cells in a chain, and may or may not produce capsules depending on the strain. It ferments dextrose, levulose, lactose, maltose, sucrose, dextrin, starch, galactose, and salicin; it does not ferment arabinose, adonitol, dulcitol, erythritol, inositol, mannitol, and sorbitol. Lactose in milk is not sufficiently fermented to produce coagulation. This organism is apparently most pathogenic for chickens and is also capable of

producing disease in rabbits, pigeons, ducks, turkeys, lambs, and white mice. Guinea pigs are highly resistant to this type of infection. *Streptococcus galli-narum* may be cultivated on agar medium to which blood has been added. Young colonies are white and gelatinous but become brownish with bluish margins with increasing age.

**Transmission.** The exact mode of the transmission of this infection is not definitely known at the present time. It is believed that contamination of feed and water may be an important factor. Hudson (1933) considered the possibility of establishing the infection in flocks through the introduction of carrier birds. Infection through the respiratory tract rather than the alimentary canal by means of contaminated food and water was also suggested.

. **Symptoms.** Peracute cases frequently die without showing definite clinical symptoms. Extreme depression and ruffled feathers may be observed in less acute cases. The most common symptoms characteristic of chronic infections include depression, ruffled feathers, somnolence, incoordination of the head, neck, legs, and wings, followed by progressive paralysis and death. Frequently cyanosis of comb and wattles develop prior to death. Diarrhea may or may not be present, and diffuse bile-stained excreta are sometimes voided prior to the development of paralysis.

**Lesions.** Peracute cases may show little or no gross pathological changes. Acute cases usually show hemorrhagic discoloration of the breast and enlargement of the liver, spleen, and kidneys. Hemorrhagic enteritis and a serofibrinous exudate in the peritoneal cavity are common findings. The lungs may contain some areas of congestion, and the heart muscle may show hemorrhages. Bushnell and Twiehaus (1939) recorded a profuse exudate and an abnormal amount of fluid around the brain and spinal cord. Hutyra and Marek (1938) described advanced cases of chronic streptococcosis showing enteritis and emaciation.

**Mortality.** Mortality may exceed 50 per cent in acute cases with little or no recovery of the affected birds, while in the less acute cases mortality may be considerably less.

**Diagnosis.** Diagnosis can be made only by the isolation of the organism. In acute cases, the streptococci may be found abundantly in the blood and parenchymatous organs and may be seen in stained smears. In chronic infections, the organism may be isolated from exudates and the tissues involved.

**Control and treatment.** Rigid sanitary practices should be used to prevent infection. Care should be taken in the introduction of new adult stock into the flock because of the possibility of introducing carrier birds. Prompt removal of the affected birds and their proper disposal is advised in an outbreak of this disease. This should be followed by thorough cleaning and dis-

infecting of the premises and equipment. No effective medicinal treatment is known at the present time.

## REFERENCES

*Streptococcosis*

Beaudette, F. R., and Hudson, C. B.: 1931. New Jersey station hints to poultrymen. 19:4.

Bergman, A. M.: 1907. Cited by Magnusson (1910).

Bushnell, L. D., and Twiehaus, M. J.: 1939. Poultry diseases, their prevention and control. Kan. Agr. Exper. Sta., Bul. 284:55.

Dammann, C., and Manegold, O.: 1905. Die Schlafkrankheit der Hühner. Deutsch. tierärztl. Wochenschr. 13:577.

Edwards, P. R.: 1933. Further studies on the differentiation of human and animal strains of hemolytic streptococci. Jour. Bact. 25:527.

——: 1934. Characters of hemolytic streptococci isolated from pathological conditions in fowls. Jour. Comp. Path. and Therap. 47:152.

—— and Hull, F. E.: 1929. Bacillary white diarrhea and related diseases of chickens. Ky. Agr. Exper. Sta., Bul. 296:237.

—— and Hull, F. E.: 1937. Hemolytic streptococci in chronic peritonitis and salpingitis of hens. Jour. Am. Vet. Med. Assn. 91:656.

Genest, P., and Nadeau, J. D.: 1944. Observations, chez la poule, d'une épizootie due á Streptococcus zooepidemicus. Canad. Jour. Comp. Med. 8:342.

Gray, E.: 1939. Further observations on fowl paralysis. Vet. Record 51:754.

Greve, L.: 1908. Beitrag zur Kenntnis der Streptokokken-Krankheit (Schlafkrankheit) der Hühner. Deutsch. tierärztl. Wochenschr. 15:213.

Gwatkin, R.: 1946. Interpretation of original work of Genest and Nadeau (1944). Vet. Bul. 16:1.

Hodosy, J.: 1944. (Streptococcosis in young geese and ducks.) Allatorv. Lapok, p. 37. (Abst. Vet. Bul. 16:426.)

Hudson, C. B.: 1933. A specific infectious disease of chickens due to a hemolytic streptococcus. Jour. Am. Vet. Med. Assn. 82:218.

Hutyra, F., and Marek, J.: 1938. Streptomycosis of fowls. In Special Pathology and Therapeutics of the Disease of Domestic Animals. Fourth Ed., Vol. I. Alexander Eger, Chicago, Ill. P. 120.

Kernkamp, H. C. H.: 1927. Idiopathic streptococcic peritonitis in poultry. Jour. Am. Vet. Med. Assn. 70:585.

Magnusson, H.: 1910. Ueber eine für Europa neune Hühnerseuche. Apoplektische Septikämie der Hühner. Zentralbl. f. Bakt., Abt. I, Orig. Bd. 56:411.

Merchant, I. A.: 1916. *Streptococcus gallinarum*. In Veterinary Bacteriology. Third Ed. Iowa State College Press, Ames, Iowa. P. 260.

Nörgaard, V. A., and Mohler, J. R.: 1902. Apoplectiform septicemia in chickens. U.S.D.A., Bur. An. Ind., Bul. 36.

Sherman, J. M.: 1937. The streptococci. Bact. Rev. 1:3.

Ward, A. R., and Gallagher, B. A.: 1920. Diseases of Domesticated Birds. Macmillan Company, New York. P. 28.

## AVIAN STAPHYLOCOCCOSIS

Avian staphylococcosis is a disease of young domesticated fowl characterized chiefly by acute septicemia or chronic arthritis. Additional manifestations may include vesicular dermatitis (Hoffman, 1939), omphalitis (Jungherr and Plastridge, 1941), keel cysts (Van Ness, 1946), and bumble foot (Gibbs, 1936).

**History.** The earliest description of the disease is credited to Prahl (1873), who described a condition in geese comparable to staphylococcus infection. Apparently the first actual incrimination of the staphylococcus organism came from Lucet (1892) of France who isolated *Staphylococcus aureus* from geese.

Other early descriptions include those by Freese (1907), Eber (1921), Hasenkamp and Sachweh (1914), Schlegel (1922), and Reinhardt (1922), all of Germany, and van Heelsbergen (1919), of Holland. Krausz (1901), of Hungary, reported a disease in chickens from which he isolated *Staphylococcus pyogenes*. He noted an extremely high mortality, but was unable to reproduce the disease. Some doubt exists as to staphylococcus infection being the primary cause. Hole and Purchase (1931) described a disease of pheasants in England in which both peracute septicemia and chronic arthritis were observed. They were able to isolate *Staphylococcus aureus* and also *Staphylococcus citreus* in rare cases.

Kawashima and Nakamura (1938) reported a disease in Japan among six- to nine-week-old chicks in which marked edema of the wing, with blood-stained serous fluid, was found. Staphylococci were isolated in every case, and artificial infection by subcutaneous inoculation was successful. In two cases *Clostridium septicum* was isolated, which they believed to be a saprophyte.

Hoffman (1939) described an outbreak of vesicular dermatitis among chickens in California involving the unfeathered integument of the head, feet, and shanks. He isolated staphylococci consistently from lesions and reproduced vesicle formations in twelve of twenty-nine inoculations on scarified skin. Inoculations were also made on scarified skin of chickens, using staphylococci obtained from a case of human impetigo, and lesions indistinguishable from those produced by staphylococci of avian origin developed.

Jungherr and Plastridge (1941) observed an outbreak of staphylococcosis in five-month-old cockerels. Anti-pick devices had been placed on the birds previous to the outbreak, and it was believed that this was a contributing factor in the spread of the disease. Mortality was slightly more than 50 per cent.

Gwatkin (1940) reported an outbreak in Canada of staphylococcus infection in mature Barred Plymouth Rock males. He described emaciation, anemia, anorexia, and severe swelling of the joints with a morbidity rate of 45 per cent of the flock. The mortality was 50 per cent of the affected birds.

Rowlands and Smith (1945) described the first recognized outbreak of staphylococcus infection of geese in Great Britain. Typical symptoms of arthritis with abscesses of the joint capsules and tendon sheaths, together with periarticular lesions, were observed.

Van Ness (1946) isolated *Staphylococcus citreus* in pure culture from keel cysts and livers of affected fowl. Arthritis was a general symptom of infected birds. He considered that injury to the keel produced the primary focus of infection which was followed by arthritis and septicemia.

**Distribution.** Cases have been reported from Europe, Japan, Canada, and the United States, indicating a worldwide distribution.

**Etiology.** *Staphylococcus aureus* has been incriminated as the causative organism in the majority of cases reported. Hole and Purchase (1931) and Van Ness (1946) succeeded in isolating *Staphylococcus citreus*. Gibbs (1936) reported *Staphylococcus albus* as a common cause of bumble foot, infection of the infraorbital sinuses, swollen wattles, pyemia, and septicemia. *Staphylococcus aureus* is commonly found in the bacterial flora of apparently normal animals. Gibbs (1931) isolated this organism from the respiratory tract of four out of ten healthy control birds. Merchant (1946) refers to *Staphylococcus aureus* as the "opportunist type" of organism because it is commonly present, awaiting suitable conditions for assuming pathogenicity. *Staphylococcus aureus* is a Gram-positive organism, does not produce spores, capsules, or flagella, and is one of the more resistant staphylococci.

**Pathogenicity.** According to Hutyra and Marek (1938), staphylococcosis occurs in young geese, ducks, and chickens, but it is not transmitted from one bird to another. Hole and Purchase (1931) reported the disease as being comparatively rare in birds.

**Symptoms.** Acute cases of staphylococcosis are often characterized by severe arthritis and some diarrhea. There may or may not be an elevation of temperature (Hutyra and Marek, 1938). Swellings of the joints are usually hot and tender. Death frequently follows in 2 to 4 days after the appearance of the initial symptoms. Chronic infections are usually manifested by a disturbance in locomotion. Swellings of the tarsometatarsal and sometimes alar joints, together with the plantar swelling, commonly occur. These swellings may at first be quite fluctuating but later become more firm and localized. Affected birds assume sitting positions on their hocks and move only with considerable effort. According to Hutyra and Marek (1938), recovery may take place in two weeks. Frequently this infection results in ankylosis of the joints. In other cases, birds die of cachexia after several weeks.

Hoffman's (1939) observations on dermatitis revealed severe vesicle formations on the unfeathered portions of the integument of the head, shanks, and feet. Van Ness (1946) observed numerous keel cysts together with arthritis. Hole and Purchase (1931) are of the opinion that birds may die of peracute septicemia, showing no symptoms of the joint involvement.

**Pathology.** The tibiometatarsal, femorotibial, and metatarsal joints are most frequently involved. The integument over and in the vicinity of the affected articulations may show superficial ulcerations. These changes have been compared with those of decubital sores (Jungherr, 1933). The swelling

of the tibiotarsal joint is produced by a serous exudate between the articular ligaments and the overlying integument, the latter also being swollen. Fibrinous deposits appear on the subcutis and the periarticular ligaments together with hemorrhagic areas. The pathological process also involves the tendinous sheaths and muscle fasciae. When the exudate becomes inspissated, the swelling is more firm. Vesicle formation of the integument of the head, comb, and wattles may appear, which is later replaced by a brownish colored crust or covering.

**Diagnosis.** Hutyra and Marek (1938) considered the simultaneous occurrence of arthritis in many fowl as pathognomonic of staphylococcosis because arthritis due to other types of infections is relatively rare. Isolation of the staphylococci from affected birds is necessary to confirm the diagnosis.

**Control.** Sharp objects and rough materials which might cause mechanical injury to the fowl should be removed from the premises. The histories of most infections indicate an initial injury of the fowl that provides an atrium of infection for the staphylococci. Prompt removal and proper disposal of all affected birds, followed by rigid sanitation, are recommended.

**Treatment.** Autogenous vaccines appear to be of no value either as a protective or therapeutic agent, according to the report of Rowlands and Smith (1945). These authors note that penicillin is definitely bacteriostatic to the staphylococci in vitro while sulfadiazine is not effective.

## REFERENCES

*Staphylococcosis*

Eber, A.: 1921. Seuchenhafte Staphylokokkenkrankheit (ansteckende Knocken-und Gelenkentzündung) des Geflügels. Deutsch. tierärztl. Wochenschr. 29:119.

Freese: 1907. Ueber eine durch den *Staphylococcus pyogenes aureus* hervorgerufene Osteo-Arthritis beim jungen Gänsen und Enten. Deutsch. tierärztl. Wochenschr. 15:322.

Gibbs, C. S.: 1931. Saprophytic and secondary microorganisms occurring in the respiratory tracts of domestic fowls and chickens in health and in disease. Jour. Bact. 21:97.

———: 1936. A comparative study of human and avian strains of *Staphylococcus albus*. Jour. Bact. 31:81.

Gwatkin, R.: 1940. An outbreak of staphylococcal infection in Barred Plymouth Rock males. Canad. Jour. Comp. Med. 4:294.

Hasenkamp and Sachweh: 1914. Staphylokokken-Erkrankungen beim Geflügel. Tierärztl. Rundschau 20:85. (Cited by van Heelsbergen, 1929.)

Hoffman, H. A.: 1939. Vesicular dermatitis in chickens. Jour. Am. Vet. Med. Assn. 95:329.

Hole, N., and Purchase, H. S.: 1931. Arthritis and periostitis in pheasants caused by *Staphylococcus pyogenes aureus*. Jour. Comp. Path. and Therap. 44:252.

Hutyra, F., and Marek, J.: 1938. Staphylomycosis of fowls. In Special Pathology and Therapeutics of the Diseases of Domestic Animals. Fourth Ed., Vol. I. Alexander Eger, Chicago, Ill. P. 121.

Jungherr, E.: 1933. Staphylococcal arthritis in turkeys. Jour. Am. Vet. Med. Assn. 82:243.

——— and Plastridge, W. N.: 1941. Avian staphylococcosis. Jour. Am. Vet. Med. Assn. 98:27.

Kawashima, H., and Nakamura, N.: 1938. Ueber eine neue durch Staphylokokkus pyogenus citrius hervorgerufene odematose Krankheit der Hühnerküchen. Jour. Jap. Soc. Vet. Sci. 17:118. (In German: Japanese summary pp. 291–92 of pt. I.) (Abst. in Vet. Bul. 12:523.)

Krausz, A.: 1901. Ueber eine bisher nicht beschriebene Hühnerepizootie. Zentralbl. f. Bakt., I. Orig. 29:980.

Lucet, A.: 1892. De l'ostéo-arthrite aiguë infectieuse des jeunes oies. Ann. Inst. Past. 6:841.

Merchant, I. A.: 1946. The genus staphylococci. In Veterinary Bacteriology. Third Ed. Iowa State College Press, Ames, Iowa. P. 276.

Prahl, T.: 1873. Krankheiten des Geflügels. Mittl. a. d. thierärztl. Praxis im Preuss Staate. 20:168. (Berichtsjahr 1871–1872.)

Reinhardt, R.: 1922. Seuchenhafte Staphylokokkenkrankheit bei Gänsen. Monatschr. f. Tier-heilk. 33:257.

Rowlands, W. T., and Smith, H. W.: 1915. Staphylococcosis in geese. Jour. Comp. Path. and Therap. 55:125.

Schlegel, M.: 1922. Staphylomykosis bei Huhn und Ente. Arch. f. wiss. Tierheilk. 47:397.

van Heelsbergen, T.: 1919. Gemengde staphylococcenen coliinfectie bij eenden. Zentralbl. f. Bakt. I. Ref. 68:272.

——: 1929. Handbuch der Geflügelkrankheiten und der Geflügelzucht. Ferdinand Enke. Stuttgart. P. 203.

Van Ness, G.: 1946. *Staphylococcus citreus* in the fowl. Poultry Sci. 25:617.

# ARTHRITIS CAUSED BY *BACTERIUM ARTHROPYOGENES, ESCHERICHIA VENEZUELENSIS,* AND *SALMONELLA PULLORUM*

*Bacterium arthropyogenes, Escherichia venezuelensis,* and *Salmonella pullorum* have been isolated from cases of chronic arthritis in fowl. These forms of arthritis are relatively rare and have a low mortality.

**History.** Nobrega (1940) described an outbreak of arthritis in Brazil among fifteen birds. He isolated *Bacterium arthropyogenes* from five of the affected birds. Joint lesions were reproduced by injection of culture directly into the joint and by intravenous inoculation. Gallo (1942) found a disease condition similar to the one described earlier by Nobrega (1940), but the causal agent was found to be *Escherichia venezuelensis.* Swellings occurred in the leg joints and plantar surfaces of the foot. He also was able to transmit the infection by experimental inoculation. Investigators in the United States have not reported arthritis caused by these two organisms.

**Etiology.** *Bacterium arthropyogenes* and *Escherichia venezuelensis* are both Gram-negative, nonsporeforming organisms. Topley and Wilson (1938) describe *Bacterium arthropyogenes* as a rod measuring 1.5μ by 0.5–0.7μ; it is noncapsulated and nonmotile. The organism stains readily with aniline dyes and is not acid-fast. It is both aerobic and facultatively anaerobic and grows well on laboratory media. Streptobacillary forms may occur when the organism is grown in broth. *Salmonella pullorum* was isolated by Beaudette (1936) from a chick showing fluctuating swellings of the tibiometatarsal joints. Reis (1942) also isolated *Salmonella pullorum* from one case of arthritis in a mature fowl.

**Pathogenicity.** The disease thus far has been found only in chickens. Gallo (1942) inoculated pigeons and other birds, which recovered. When *Bacterium arthropyogenes* is inoculated directly into the tibiotarsal joint, using a 48-hour broth culture, severe arthritis may develop in approximately 3 days. The lesion soon becomes suppurative. Death frequently occurs in 5 to 25 days. Inoculation into the wing vein is followed by a severe arthritis of the tibiotarsal joints in 3 to 5 days, death occurring within an average of 13 days. Kidney, liver, and pericardial lesions may appear when the organism is inoculated intravenously.

**Symptoms.** Lameness, inappetence, and painful semifluctuating swellings of the tibiotarsal and metatarsal joints are common symptoms. These swellings may result in abscess formation, sometimes involving the musculature and tendinous insertions of the affected tibiotarsal joints (Fig. 16.1 and 16.2).

**Lesions.** Large abscesses may be found in and around the joints, and in some cases microscopic foci appear in the liver. Perivascular infiltrations have been found in the lungs and spleen.



Fig. 16.1. Periarticular abscesses caused by *B. arthropyogenes*. Spontaneous case. Courtesy P. Nobrega, Arq. do Inst. Biol., São Paulo.

**Transmission.** Symptoms and lesions may be produced by inoculation directly into the joint cavity, but oral administration of the causative organism does not produce the disease (Nobrega, 1940). The natural mode of infection is not known.

**Recovery.** Nobrega (1940) observed retrogression of symptoms in some birds after 20 to 30 days.

### REFERENCES

*Arthritis*

Beaudette. F. R.: 1936. Arthritis in a chick caused by *Salmonella pullorum*. Jour. Am. Vet. Med. Assn. 89:89.

Gallo, P.: 1942. Estudios sobre una nueva entidad nosográfica: "La artritis infecciosa de las aves, y su agente": "La Escherichia Venezuelensis," n. sp. Rev. Med. vet. Parasit., Caracas. 4:3. (Abst. Vet. Bul. 17:357.)

Nobrega, P.: 1940. Artrite em galinha, produzida por "*Bacterium arthropyogenes*." Arq. Inst. Biol. São Paulo. 11:323.

Reis, J.: 1942. Artrite em galinha produzida por *Salmonella pullorum*. Arq. Inst. Biol. São Paulo. 13:115.

Topley, W. W. C., and Wilson, G. S.: 1938. The Principles of Bacteriology and Immunity. Second Ed. William Wood and Company, Baltimore, Md. Pp. 1183, 1193.

## AVIAN COLIBACILLOSIS

*Escherichia coli* is frequently isolated from fowl, but its status as a patho-
gen has not been clearly established. The presence of the organism in both
young and adult fowl has been associated with acute and chronic disease
conditions.

**History.** Lignières (1894), Martel (1897), Claussen (1907), and Zeiss
(1914) were among the first to report septicemic infections caused by
*Escherichia coli*. Lignières (1894) isolated *Escherichia coli* from affected



Fig. 16.2. Plantar abscess produced by *B. anthropyogenes*. Spontaneous case. Courtesy
P. Nobrega, Arq. do Inst. Biol., São Paulo.

birds and inoculated the organism into hens which resisted subcutaneous and
intramuscular injections of 1 to 2 cc. of culture. The pigeon succumbed to
a dosage of 1 cc. of the same culture. Martel (1897) described colibacillosis
of chickens and turkeys. He killed hens with small amounts of culture by
intramuscular injection but failed to cause infection by intravenous inocu-
lation or by the introduction of cultures, excrement, and virulent products
into the alimentary tract. Claussen (1907) isolated a strain of *E. coli* which
was uniformly fatal to canary birds, white mice, and guinea pigs, but which
affected hens, ducks, and pigeons only in exceptional cases. He suggested
that coliform organisms normally found in the digestive tract possess the
ability of enhancing their virulence under conditions of fatigue and other

debilitating influences after which the organisms invade the body, producing a fatal septicemia. Zeiss (1914) reported a fatal septicemia in two hens. Autopsy revealed softening of the liver and spleen ·with some subcutaneous hemorrhages. This investigator isolated an organism which he designated as *Bacterium coli*. The organism was pathogenic for canaries but failed to infect guinea pigs. Hempel (1913) described an outbreak of colibacillosis in which 70 of 100 chicks died within a period of three weeks. He isolated the organism from three of six chicks examined.

Colibacillosis of pigeons was first described by Sanfelice (1895). De Blasi (1906) also observed coliform organisms in pigeons and isolated pure cultures from the blood, fibrinous exudates of the liver, and pericardium. He reproduced colibacillosis by experimental inoculation. Fiorentini (1896) described the disease among swans and geese in Milan. Zander (1923) attributed an epizootic among young geese in East Prussia chiefly to *E. coli*. Spissu (1907) and van Heelsbergen (1929) isolated coliform organisms from ducks.

Bueno (1940) studied the distribution of the coliform aerogenes group of organisms in fowl. He classified the findings on the basis of Parr's "IMVIC"[1] test (Parr, 1938) and found bacteria of the coli group to be common, especially in the lower digestive tract. Garrard (1946) reported on the coliform contamination of 1,080 eggs from hens infected with *Salmonella pullorum*. Coliform organisms were recovered from 78 eggs. Later, a similar study was made on 1,000 eggs from pullorum-free hens which failed to yield a single coliform organism. Osborne, Witter, and Hitchner (1946) compared cultures of microorganisms involved in chronic colibacillosis of fowls. Six strains were studied, five of which were isolated from adult fowl and one from a duck. All of the birds showed cholera-like symptoms. Inoculations into young chicks of 1 cc. intraperitoneally resulted in illness in 24 hours. Death occurred in only two of the 24 inoculants, the remainder recovering in approximately three weeks. Bushnell and Twiehaus (1939) associate colibacillosis with heavy losses in chicks of two to eight weeks of age. Improper feeding, brooding, and sanitation are given as predisposing factors to infection. *Escherichia coli* (*Bacillus coli*, *Bacterium coli*) has been isolated in pure culture from suspected cases; however, doubt still remains as to its actual role in producing disease. Some investigators suggest that there is a probability of another factor which makes the bird susceptible, preparing the way for *E. coli* migration into the blood stream, and that the prolific growth of the *E. coli* masks the true pathogen. Hutyra and Marek (1938) cite experiences wherein pure cultures of *E. coli* were isolated, but in all cases other primary causes of death were found. The work of Bueno (1940)

_____
[1] Indol, methyl red, Voges-Proskauer, and citrate reactions.

and Garrard (1946) indicate that the coliform organism is commonly found in apparently healthy birds.

**Pathogenicity.** Colibacillosis has been reported in chickens, turkeys, pheasants, canaries, geese, ducks, swans, grouse, and quail as well as other wildlife birds. It affects both the young and adult of the avian species.

**Symptoms.** The disease is usually acute and less often chronic (Hutyra and Marek, 1938). In the acute cases affecting young chicks, only mild symptoms may be noted. The symptoms are much like those of pullorum disease in the more chronic cases involving young chickens (Bushnell and Twiehaus, 1939). Droopiness, inappetence, and dyspnea are often encountered. Adult fowl affected with acute septicemia may show symptoms indistinguishable from those of fowl cholera. Depression, inappetence, ruffled feathers, dyspnea, diarrhea, and in the late stage, cyanosis, are common symptoms. Birds with subacute and chronic infections may linger in an unthrifty state for a week or more.

**Lesions.** Acute cases of septicemia in adult fowl may be characterized by inflammation of the intestine with serous hemorrhages, serofibrinous peritonitis, hypertrophy, and softening of the spleen, and hyperemia with slight swelling of the liver. The pericardium may show inflammation and the presence of hemorrhages. Pneumonia with considerable consolidation and some fibrinous adhesions to the thoacic wall are noted in chronic cases.

**Diagnosis.** Isolation of the organisms from the blood and parenchymatous organs is necessary for an accurate diagnosis.

**Control.** Overcrowding, dampness, and filthy environment must be corrected. Sick birds should be removed promptly and properly disposed of to prevent further spread of the disease.

## REFERENCES

*Colibacillosis*

Bueno, R. C.: 1940. Identificacao do grupo coli-aerogenes em aves. Distribuicao e frequencia. Arq. Inst. Biol., São Paulo. 11:69.

Bushnell, L. D., and Twiehaus, M. J.: 1939. Poultry diseases, their prevention and control. Kan. Agr. Exper. Sta., Bul. 284:63.

Claussen, L.: 1907. Über Kolibakterienseptikämie bie Hühnern als Transportkrankheit. Zeitschr. f. Infektionskr. d. Haustiere 3:69.

De Blasi: 1906. Colibacillosis. Jahresbr. Vet. Med. 26:350.

Fiorentini, A.: 1896. Hamorrhagische Septikämie der Schwane. Zentralbl. f. Bakt., I. Orig. 19:932.

Garrard, E. H.: 1946. Coliform contamination of eggs. Canad. Jour. Res. 24:121.

Hempel, H.: 1913. Über eine Colibacillose der Hühner. Inaugural Dissert., Hannover. (Cited by van Heelsbergen, 1929.)

Hutyra, R., and Marek, J.: 1938. Colibacillosis of fowls. In Special Pathology and Therapeutics of the Diseases of Domestic Animals. Fourth Ed., Vol. I. Alexander Eger, Chicago, Ill. P. 119.

Lignières, M. J.: 1894. Septicémie à coli-bacille chez la poule. Compt. rend. Soc. de biol. 46:135.

Martel, M.: 1897. Maladie à coli-bacille de la poule et de la dinde. Compt. rend. Soc. de biol. 49:500.

Osborne, J. C., Witter, J. F., and Hitchner, E. J.: 1946. A comparative study of cultures of microorganisms involved in chronic colibacillosis in fowl. Mich. St. Coll. Vet. 6:25.

Parr, L. W.: 1938. The occurrence and succession of coliform organisms in human feces. Am. Jour. Hyg. 27:67.

Sanfelice, F.: 1895. Eine Seuche bei Tauben durch *Bacterium coli* verursacht. Zeitschr. f. Hyg. 20:23.

Spissu: 1907. Colibacillose bei der Ente. Jahresbr. Vet. Med. 27:334.

van Heelsbergen, T.: 1929. Handbuch der Geflügelkrankheiten und der Geflügelzucht. Ferdinand Enke, Stuttgart. P. 199.

Zander, M.: 1923. Untersuchungen über ein seuchenartiges Sterben der Gänsekücken im Jahre 1922. Inaugural Dissert., Berlin. (Cited by van Heelsbergen, 1929.)

Zeiss, H.: 1914. Koliseptikämie bei Hühnern. Arch. f. Hyg. 82:27.

## HJÄRRE'S DISEASE
### (Coli-granuloma, Colibacillosis)

Hjärre and Wramby (1945) describe tuberculosis-like granulomae in adult fowl from which they isolated a mucoid form of coli organism. The organism is Gram-negative and capsulated but is not acid-fast. It develops mucoid colonies on plate cultures. By using the M form of the organism, Hjärre and Wramby produced the disease in experimental fowl. Both intramuscular injections of tissue pulp from the granulomae and intravenous injections of pure culture were used in transmission experiments. The S and R forms were less pathogenic for fowl than the M form. Both white mice and rabbits also proved susceptible to the organism. However, attempts to infect fowl by feeding pure cultures failed to produce the disease.

Eighteen strains of the organism have been divided into five fermentation types related also to serological types. Intraportal inoculation of mucoid coli organisms into fowl was followed by the appearance of typical coligranulomae within one week, whereas the introduction of avian tubercle bacilli did not result in the appearance of typical tubercles until 20 days later. Considerable differences are noted in the early stages of the two types of granulomae, chiefly in the size and appearance of the giant cells. However, the older coli-granulomae resemble tubercles and cannot be distinguished morphologically. Coli-granulomae are most commonly found in the ceca and liver, but they also occur in the proventriculus, large and small intestines, spleen, bone marrow, and lungs. The granulomae caused by this mucoid or M form of coli organism are softer than tubercles, the former tending to regress.

The disease appears to be only slightly infectious under natural conditions, but its importance is based chiefly on the similarity of the lesions with those of tuberculosis.

### REFERENCE

*Hjärre's Disease*

Hjärre, A., and Wramby, G.: 1945. Undersökningar över en med specifika granulom förlöpande hönssjukdom orsakad av mukoida kolibakterier (Koli-granulom). Skand. Veterinärtidski. 35:449.

# DISEASES CAUSED BY FUNGI

By K. L. BULLIS, *Department of Veterinary Science, University of Massachusetts, Amherst, Massachusetts*

☆    ☆    ☆

## ASPERGILLOSIS

Aspergillosis has been observed in many birds and mammals. Frequent reference is made to the relationship of the disease in man to occupation, particularly in the so-called *graveurs des pigeons* (pigeon feeders).

**Occurrence.** Aspergillosis is encountered in poultry in two main forms. Acute outbreaks in which there is a high morbidity and a high mortality may occur particularly in young birds. In adults especially, an occasional bird in a flock or aviary may become affected while the other birds remain healthy. The numerous reports in the literature suggest that nearly all species of birds may be affected. The incidence of the disease is not great, however, as evidenced by reports from diagnostic laboratories.

**Etiology.** It is generally agreed that *Aspergillus fumigatus* Fresenius is the most pathogenic and the most frequently encountered in disease processes due to aspergilli. The spores are widely distributed in nature, and birds frequently come in contact with them through contaminated feed or litter. The fungus grows quite readily on the ordinary laboratory culture media at room temperature, at 37° C., and higher. Czapek's solution agar or Sabouraud's agar may be used. The colonies are green to bluish-green at first and darken with age so as to appear almost black. The colonies vary from velvety to floccose. The conidiophores are short, up to 300μ long by 2 to 8μ in diameter, the vesicles are apical flask-shaped up to 20 to 30μ in diameter, the sterigmata are 6 to 8μ by 2 to 3μ, and the conidia are globose 2.5 to 3μ in diameter, in chains forming solid columns up to 400μ by 50μ (Fig. 17.1) (Thom and Church, 1926).

Leber, according to van Heelsbergen (1929) and Lucet (1897), succeeded in isolating toxins from cultures of *Aspergillus fumigatus.* Ceni and Besta (1902) were able to extract toxic materials from spores. A toxin reported by Bodin and Gautier (1906) was similar to bacterial toxins and produced clonicotonic convulsions, paralytic symptoms, and finally death. A toxin obtained by Henrici (1939) was toxic for rabbits, guinea pigs, mice, and chickens. This toxin was hemotoxic, neurotoxic, and histotoxic. Rabbits and dogs are very susceptible to Aspergillus toxin. Pigeons, however, which

are very susceptible to spontaneous infection are very resistant to injected toxin.

Vigorous, healthy birds apparently can withstand considerable exposure to Aspergillus spores occurring under natural conditions. Inhalation of a considerable number of spores, as may occur when the litter or feed are heavily contaminated, may result in infection. The occasional bird which becomes infected in a flock which is otherwise healthy, may do so because of lowered resistance or severe individual exposure. Aspergillosis can readily be produced experimentally by intrathoracic injection in chickens and pigeons. Schütz (1884), Bollinger, cited by van Heelsbergen (1929), and others observed that infection of the lungs was established following inhalation of spores. Walker (1915) reported that 5- to 7-day-old ostriches succumbed in 2 to 8 days to aspergillosis in the lungs and air sacs if spores were blown into the trachea. Intravenous inoculation resulted in pulmonary and hepatic aspergillosis. Young ostriches also developed the disease when kept on straw which had been artificially contaminated. Durant and Tucker (1935) produced the disease in a poult by feeding mash from which *A. fumigatus* was isolated.
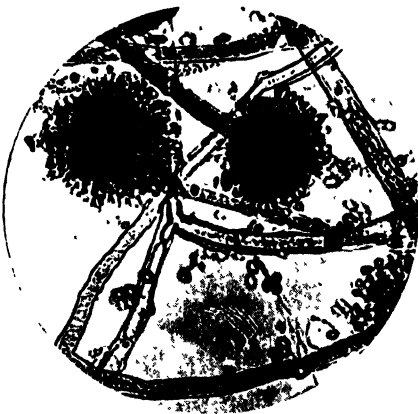


FIG. 17.1. *Aspergillus fumigatus.* X250. (From Nowak: Documenta Microbiologica, courtesy Gustav Fischer.)

*Aspergillus glaucus* and *A. niger* may be encountered in some cases, particularly in cutaneous lesions. Lahaye (1928) discusses cutaneous aspergillosis in pigeons. Mucor sp. and Penicillium sp. and other fungi may be encountered in pulmonary mycosis, particularly in mixed infections (Baker, Courtenay-Dunn, and Wright, 1934; Thompson and Fabian, 1932).

**Lesions.** The lesions depend considerably on the site of infection. Either localization or generalization may be observed. Individual lesions may be observed, for example, in the syrinx (Fig. 17.2) or in a single air sac. The lungs are most frequently involved. Pulmonary lesions vary from miliary nodules up to larger nodules (Fig. 17.3). In some cases there may be localized hepatization, and in others grossly visible mycelial masses may be present in the air passages and bronchi. There may be generalized involvement of the air sacs. Occasionally, a circular disc-shaped necrotic mass with a concave surface, loosely attached to which there is a circular more or less flat or convex placque, may be observed. Various manifestations of the disease have been described. Lange (1914) recorded nodules in the lungs, the thoracic, and the abdominal cavities of chickens, ducks, geese, and pigeons.

These nodules varied from pinhead or millet seed size up to the size of a pea. They were yellow in color, of an elastically soft or cartilaginous consistency, and homogeneously caseous. Individual nodules were noted on the intestinal serosa and in the parenchyma of the liver in a goose.

The nodules in the lungs of a turkey observed by Schlegel (1915) were pinhead to lentil size and were surrounded by an infiltrated or hemorrhagic corona with considerable hepatization. There were also grayish-yellow, fibrinopurulent disc- or plate-shaped masses of exudate 2 to 5 mm. thick on the pleura. Inflammation and detrital masses were present in the bronchi.
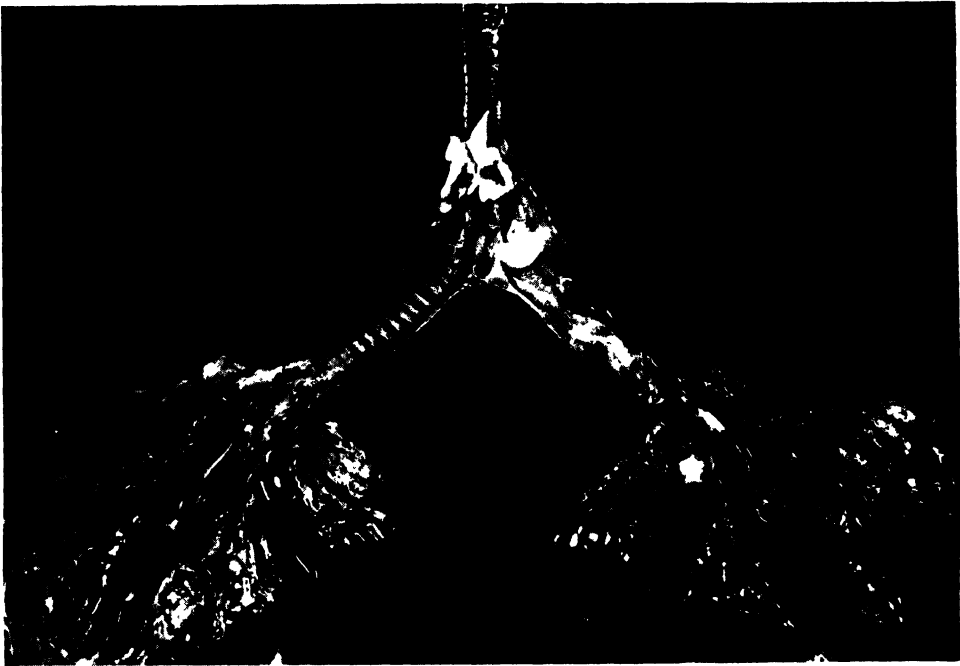


FIG. 17.2. Aspergillosis involving syrinx.

The anterior thoracic, the axillary, and the cervical air sacs contained yellow caseous flat discs and masses consisting of inflammatory exudate and mycelia. The left lower, upper posterior thoracic, and left abdominal air sacs were greatly distended. The walls of these air sacs were thickened and covered with a furlike growth of mold. Adjacent to the air sacs there were lentil-sized, knob-shaped, and concentrically layered, turbid yellow, solid nodules composed of fibrin and mycelia. There were about 200 cc. of a reddish, turbid fluid in the abdominal cavity.

There were no circumscribed yellowish foci in the outbreak in chicks reported by Savage and Isa (1933). There was a diffuse grayish-yellow infil-

tration in the lungs with about one-third of each lung involved. The rate of mortality was 90 per cent in this outbreak.

In pneumomycosis in a flamingo described by Mohler and Buckley (1904), the lungs were filled with nodules, and the mucosa of the bronchi was covered with membranous masses that consisted primarily of the fungous mycelium.

Archibald (1913) found gray, round colonies of the fungus in the bronchioles in an ostrich, whereas in a case described by Jowett (1913) the lungs were covered with miliary foci.



FIG. 17.3. Aspergillosis nodules in lungs and placque-like formations on the serous membranes.

Lahaye (1928) states that *Aspergillus glaucus* may be the cause of a disease of the skin in pigeons, particularly in young birds. Any part of the body may be affected with yellow scaly spots. The feathers in the affected areas are dry and easily broken.

Durant and Tucker (1935) observed yellowish-white nodules up to 8 × 5 mm. in the lungs of wild turkey poults being reared in captivity. The hyphae of the fungus also penetrated the tissue of the lung, and there was involvement of the adjacent air sacs.

In canaries observed by de Jong (1912) there were small whitish-yellow, crusty coatings on the tongue, palate, aditus laryngis, and in the trachea and syrinx. Caseous foci in the lungs and caseous coatings on the pleura and peritoneum were also observed.

The histological picture as described by Nieberle (1923) consists of focal pneumonia, multiple necrosis, and nodular formations which resemble tubercles. The diffuse pneumonic foci are indicative of fibrinous or catarrhal pneumonia. The alveoli, bronchioles, and bronchi are filled with mucus, stained fibrin, nuclear fragments, detritus, leucocytic and inflammatory cells,

and mycelia. The mycelia penetrate the walls, and the surrounding pulmonary parenchyma shows an exudative cellular inflammation or necrosis. The tubercle-like nodules show in the center a radiating turf of hyphae surrounded by a reactive inflammatory wall which resembles granulation tissue. Foreign body giant cells are frequently observed. The fruiting organs (conidiophores, sterigmata, conidia) occur more frequently in the air sacs.

**Symptoms.** Dyspnoea, gasping, and accelerated breathing may be present. When these symptoms are associated with other respiratory diseases such as infectious bronchitis and infectious laryngotracheitis, they are usually accompanied by gurgling and rattling noises, whereas in aspergillosis there usually is no sound. Guberlet (1923) ascribed somnolence, inappetence, emaciation, increased thirst, and pyrexia to aspergillosis. Cases under his observation emaciated rapidly and showed a diarrhea in the later stages. Dysphagia was noted in cases in which the esophageal mucosa was involved. Mortality was as high as 50 per cent in confined birds on some farms, whereas birds running out of doors were more resistant and escaped infection entirely on other farms. According to van Heelsbergen (1929) some investigators have reported serous excretions from the nasal and ocular mucosa. Extreme dyspnoea was recorded by de Jong (1912) in canaries. In an outbreak in wild turkey poults reared in captivity, described by Durant and Tucker (1935), mortality began at 5 days, reached a peak at 15 days, and subsided at three weeks of age. Some affected poults died in convulsions within 24 hours. In two lots of poults 200 survived out of 785. Gauger (1941) reported an outbreak in adult chickens in which about 10 per cent of the flock was affected with symptoms not unlike those shown by birds affected with laryngotracheitis and in which there was no abnormal mortality, but the egg production was temporarily lowered. Reis (1940), cited by Hudson, and Hudson (1947) have reported infection of the eyes in chicks two to five weeks of age. Infection in Reis' cases originated in sawdust litter and in Hudson's cases in bagasse litter. The outbreaks were characterized by the formation of a yellow cheesy pellet beneath the nictitating membrane which caused the lids to bulge. There was some central ulceration of the cornea in the older chicks.

**Diagnosis.** The fungus can be demonstrated by cultural methods if it cannot be demonstrated in fresh microscopic preparations. Occasionally, masses of the fungus are visible to the naked eye in the air passages of the lungs, in the air sacs, or in the abdominal cavity. The typical fruiting heads may be readily demonstrated in such lesions. Demonstration of the fungus by direct examination may be impossible in the small caseous nodules seen particularly in the lungs.

**Prophylaxis and treatment.** The avoidance of moldy litter or feed serves to prevent outbreaks of aspergillosis. An examination of the premises or

materials used for feed or litter will usually reveal the source of the infection.

Treatment of affected individuals is usually considered useless. They should be sacrificed and the offending infective material removed. Lahaye (1928) reported favorable results in the treatment of aspergillosis of the skin in pigeons by the use of $HgCl_2$ solution 1:500. The surface of the body was moistened or the birds dipped into the solution, following which they were rinsed in lukewarm water and dried.

## REFERENCES

Archibald, R. G.: 1913. Aspergillosis in the Sudan ostrich. Jour. Comp. Path. and Therap. 26:171.

Baker, A. Z., Courtenay-Dunn, J., and Wright, M. D.: 1934. Observations on fungal pneumonia in the domestic fowl. Vet. Jour. 90:385.

Bodin, E., and Gautier, L.: 1906. Note sur une toxine produite par l'*Aspergillus fumigatus*. Ann. de l'Inst. Pasteur 20:209.

Ceni, C., and Besta, C.: 1902. Ueber die Toxine von *Aspergillus fumigatus* und *A. flavescens* und deren Beziehungen zur Pellagra. Zentralbl. f. Allg. Path. und Path. Anat. 13:930.

de Jong, D. A.: 1912. Aspergillosis der Kanarienvögel. Zentralbl. f. Bakt. I. Orig. 66:390.

Durant, A. J., and Tucker, C. M.: 1935. Aspergillosis of wild turkeys reared in captivity. Jour. Am. Vet. Med. Assn. 86:781.

Gauger, H. C.: 1941. *Aspergillus fumigatus* infection in adult chickens. Poultry Sci. 20:145.

Guberlet, J. E.: 1923. An epizootic of aspergillosis in chickens. Jour. Am. Vet. Med. Assn. 63:612.

Henrici, A. T.: 1939. An endotoxin from *Aspergillus fumigatus*. Jour. Immunol. 36:319.

Hudson, C. B.: 1947. *Aspergillus fumigatus* infection in the eyes of baby chicks. Poultry Sci. 26:192.

Jowett, W.: 1913. Pulmonary mycosis in the ostrich. Jour. Comp. Path. and Therap. 26:253.

Lahaye, J.: 1928. Maladies des pigeons et des poules, des oiseaux de bassecour et de volière. Remouchamps: Steinmetz-Haenen. 393.

Lange, W.: 1914. Schimmelpilzerkrankungen beim Geflügel. Deut. tierärztl. Wochenschr. 22:642.

Lucet, A.: 1897. Experimental and clinical study of *Aspergillus fumigatus*. Vet. Jour. 44:215–17; 285–88; 392–94; 45:226–31; 301–4. (Translated from Bul. de la Soc. Centrale de Méd. Vét.)

Mohler, J. R., and Buckley, J. S.: 1904. Pulmonary mycosis of birds with report of a case in a flamingo. U.S.D.A., Bur. An. Ind., Cir. 58:122–26.

Nieberle, K.: 1923. Die Lungenaspergillose. In Joest, E.: Spez. path. Anat. der Haustiere. R. Schoetz, Berlin. 3:804–7.

Reis, J.: 1940. Queratomicose Aspergilica Epizoótica em Pintos. Arquivos do Instituto Biologico. São Paulo, Brazil, Vol. 11, Artigo 48:437–50.

Savage, A., and Isa, J. M.: 1933. A note on mycotic pneumonia of chickens. Sci. Agr. 13:341.

Schlegel, M.: 1915. Schimmelpilzerkrankung (Aspergillose) in den Lungen bei Tieren. Berlin. tierärztl. Wochenschr. 31:25.

Schütz: 1884. Eindringen von Pilzsporen in die Atmungswege und die dadurch bedingten Erkrankungen der Lungen und über den Pilz des Hühnergrindes. Mittheil. a. d. Kaiserl. Gesundheitsamte. 2:208.

Thom, C., and Church, Margaret B.: 1926. The Aspergilli. Williams and Wilkins Co., Baltimore. 272 pp.

Thompson, W. W., and Fabian, F. W.: 1932. Molds in respiratory tract of chickens. Jour. Am. Vet. Med. Assn. 80:921.

van Heelsbergen, T.: 1929. Handbuch der Geflügelkrankheiten und der Geflügelzucht. Ferdinand Enke, Stuttgart. 312–22.

Walker, J.: 1915. Aspergillosis in the ostrich chick. Union of South Africa, Dept. Agr. Ann. Rpts., Dir. Vet. Res., 3–4, 535–74.

## FAVUS

Favus is a chronic dermatomycosis affecting chickens, occasionally turkeys and some other birds, animals, and man. In the fowl, the comb is almost always attacked, but other portions of the head may be affected and in severe cases the disease spreads to the feathered portions of the body.

**Occurrence.** Favus occurs only infrequently in the United States. Possibly

this is due to the lesser number of the heavier Asiatic breeds which are reported to be more susceptible. The disease is reported to be quite common in France.

**Etiology.** The causative fungus in the fowl is *Achorion gallinae*. Cultivation on Sabouraud's glucose agar is satisfactory. It is sometimes of assistance to cover the inoculum with absolute alcohol and evaporate the alcohol to destroy the contaminating bacteria. Cultures grow slowly, though more rapidly than does the type species of Achoria, *Achorion schoenleini*. Colonies develop as small round discs which are white and velvety and have small central cups and radial grooves. A reddish pigment varying from rose or strawberry red to a deep raspberry diffuses through the medium. Microscopically, the branched mycelium is twisted, the septa are irregularly spaced, the spores are in clusters, and there are nodular organs and fuseaux. Typical lesions may be produced by inoculation of the scarified comb. Mice, rabbits, or guinea pigs may also be inoculated, although the lesions are not so typical in the guinea pigs (Dodge, 1935; Jacobson, 1932).



Fig. 17.4. Favus of comb and wattles.

**Symptoms and lesions.** Lesions usually develop first on the comb. As the fungus spreads, white spots develop, the surface of which scales off, and the comb may appear as though sprinkled with flour (Fig. 17.4). Young birds with well-developed combs are most likely to be affected. The wattles and unfeathered portions of the head may be affected. As the disease progresses, the scaly deposits become thicker and form a wrinkled crust. Spontaneous recovery is reported in some cases. In other cases the fungus spreads to feathered portions of the body. The feathers fall out in patches. The skin becomes thickened in the affected areas and covered with scales and crusts, especially around the feather follicles. A moldy odor may be detected. Matruchot and Dassonville (1899) reported the appearance of favus simultaneously on the feathered and nonfeathered portions of the body. Symptoms were not extensive in the cases observed by Sabouraud, Suis, and Suffran (1909). Schlegel (1909) reported depression, weakness, emaciation, anemia, cachexia, and icterus in affected chickens In some birds, in addition to the external lesions, there were necrotic foci, nodules, and yellowish caseous deposits on the mucosa of the upper respiratory and digestive tracts. Occasionally, the bronchi and lungs were affected and necrotic caseous inflammation was observed in the crop and small intestine. The favus fungus

could be demonstrated microscopically in these lesions. The fungus spreads slowly from bird to bird by direct contact and by the scales which become detached from affected individuals and contaminate the premises.

**Diagnosis.** The characteristics of the gross lesions may be sufficient for diagnosis. If this is inconclusive, the fungus can be checked microscopically and culturally. Transmission of the disease to laboratory birds or animals or a study of the contagious nature of the disease in the affected flock may be helpful.

**Prophylaxis and treatment.** Care should be exercised in the addition of new birds to the flock. Infected houses should be cleaned and disinfected. Badly affected birds should be sacrificed. Mildly affected birds should be segregated and treatment can be tried if desired. The majority of mildly affected birds will recover without treatment. Several individuals have been observed in which various treatments were used on one side of the head and the opposite side was left untreated, and recovery was similar on each side. Van Heelsbergen (1929) suggests the following remedies: iodine and glycerine (tinct. iodine 1.0, glycerine 6.0), green soap, and 5 per cent phenol solution, or bichloride of mercury (1:500), the latter to be used particularly on the body. Beach and Halpin (1918) found an ointment of formaldehyde and vaseline to be effective. This is prepared by melting vaseline in a jar in a water bath. Five per cent by weight of commercial formalin is added, the cover tightened, and the mixture shaken until the vaseline has solidified. One or two applications well rubbed into the lesions usually suffice.

## REFERENCES

Beach, B. A., and Halpin, J. G.: 1918. Observations on an outbreak of favus. Jour. Agr. Res. 15:415.

Dodge, C. W.: 1935. Medical Mycology. C. V. Mosby Co., St. Louis. 554–55.

Jacobson, H. P.: 1932. Fungous Diseases. Charles C. Thomas, Springfield, Illinois. 52–55.

Matruchot, L., and Dassonville, C.: 1899. Recherches expérimentales sur une dermatomycose des poules et sur son parasite. Rev. Gén. de Bot. 11:429.

Sabouraud, R., Suis, A., and Suffran, F.: 1909. La "crête blanche" (favus) de la poule et son parasite. Rév. Vét. 34:601, 672.

Schlegel, M.: 1909. Favuskrankheit (Hühnergrind). Berlin. tierärztl. Wochenschr. 25:689.

van Heelsbergen, T.: 1929. Handbuch der Geflügelkrankheiten und der Geflügelzucht. Ferdinand Enke, Stuttgart. 322–27.

## THRUSH (MYCOSIS OF THE DIGESTIVE TRACT)

Stomatitis oidica, muguet, soor, moniliasis, oidiomycosis, and sour crop are other terms applied to mycotic affections of the digestive tract.

**Occurrence.** Mycosis of the digestive tract probably occurs rather frequently, but in many cases it does not appear to be of sufficient significance to be considered seriously. Numerous general discussions of poultry diseases fail to mention this disorder, and the paucity of diagnoses in reports from diagnostic laboratories suggests that it may not be of great consequence.

However, serious outbreaks have been reported in many species of birds. Animals and man are also affected. Thrush has been observed in chickens, pigeons, geese, turkeys, pheasants, ruffed grouse, and quail.

**Etiology.** The etiological significance of yeastlike fungi in affections of the digestive tract of man was recognized by Langenbeck in 1839. Questions relating to the validity of species described and their generic nomenclature have retarded a proper understanding of this type of disease. Jungherr (1933b, 1934) found *Monilia albicans, Monilia krusei,* and *Oidium pullorum* n.s. to be associated with cases of thrush, but considered that *M. krusei* was not of etiological significance. Mucor sp. and Aspergilli were also found in association with some cases. Hinshaw (1933) reported that *M. albicans* was found in most cases of thrush in turkeys and chickens which came to his attention. Both investigators considered that the mycotic infections were apt to be associated with unhygienic surroundings and perhaps secondary to other debilitating conditions. Eberth (1858) and Schlegel (1912) identified organisms observed by them as *Oidium albicans.*

The studies of Worley and Stovall (1937), Benham (1931), Martin and associates (1937), and others indicate the complexity of the problem. Stovall (1939) pointed out a means of improving the present uncertain status. He suggested a specific set of environmental conditions under which the biological characteristics of the organism were constant and could be demonstrated. Jungherr's (1934) characterization is as follows: *"Monilia albicans:* It is of widespread occurrence in gallinaceous birds, pathogenic to birds and also to rabbits on intravenous injection, and is indistinguishable from strains isolated from human sources. On Sabouraud agar it produces a whitish, creamy, high-convex colony after incubation for 24 to 48 hours at 37° C. Young cultures consist of oval budding yeast cells, about $5\frac{1}{2}$ by $3\frac{1}{2}\mu$ in dimension. Older cultures show septate hyphae and occasionally spherical, swollen cells with thickened membrane, the so-called chlamydospores. In Dunham's peptone water containing 1 per cent fermentable substance and 1 per cent Andrade's indicator, the organism produces acid and gas in dextrose, levulose, maltose, and mannose, slight acid in galactose and sucrose, and does not attack dextrin (variable according to brand), inulin, lactose, and raffinose. Gelatin stab cultures show short, villous to arborescent outgrowths without liquefaction of the medium."

The term "medical monilias" is frequently used in connection with the generic term Monilia since the term Monilia is also used for a separate group of fungi.

**Symptoms and lesions.** The symptoms are not particularly characteristic. Affected chicks show unsatisfactory growth, a stunted appearance, listlessness, and roughness of the feathers. Lesions occur most frequently in the crop (Fig. 17.5) and consist of a thickening of the mucosa with whitish, circular,

raised ulcer formations, the surfaces of which tend to scale off. Pseudo-membranous patches and easily removed necrotic material over the mucosa are not uncommon. The mouth and esophagus may show ulcer-like patches. When the proventriculus is involved, it is swollen, the serosa has a glossy appearance, and the mucosa is hemorrhagic and may be covered with a catarrhal or necrotic exudate. Histologically, Jungherr (1933a) reports the crops "showed extensive destruction of the stratified epithelium deep in the Malpighian layer and quite often walled-off ulcers or extensive diphtheroid
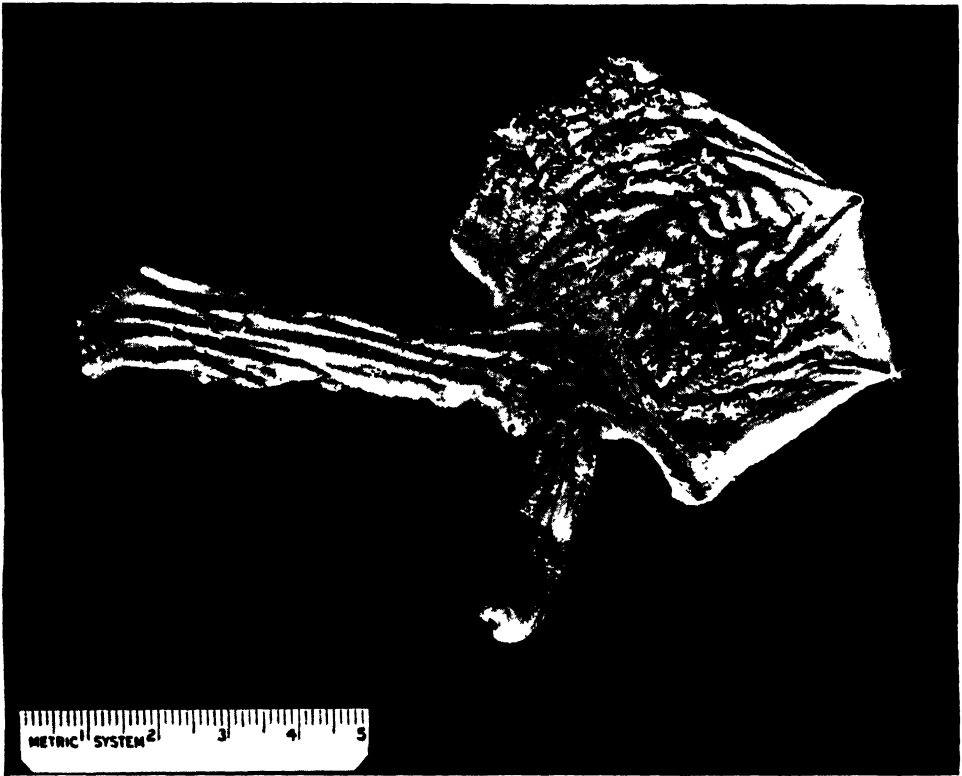


Fig. 17.5. Moniliasis in crop of turkey.

to diphtheritic membranes. The lesions were characterized by the absence of inflammatory reaction." Periportal focal necrosis in the liver in some cases suggested a toxic action upon the system.

The frequent association of mycosis of the digestive tract with other debilitating conditions such as gizzard erosions and intestinal coccidiosis must be considered. Gizzard erosions as such probably are not directly related to thrush. Likewise, the thickened intestine with watery contents frequently noted in cases of thrush is probably due to coccidiosis or other protozoan infections.

In the case of thrush reported by Eberth (1858), the esophagus, crop, and proventriculus showed an ulcerated and scaly condition. The spores and hyphae of what he termed *Oidium albicans* could be readily demonstrated in the lesions. The proventriculus was the principal organ involved in the cases observed by Schlegel (1912). The mouth, pharynx, and crop were, however, involved in some cases. Schlegel (1921) also observed the disease in geese. Diphtheroid lesions were noted in the proventriculus and small intestine. Abscess formations were present under pulpy, soft, grayish-white to brownish-red necrotic masses. Hinshaw (1933) reported thrush in twelve flocks of turkeys, and the lesions were similar to those noted in chickens. Zürn (1882) and Klee (1899) described the disease in pigeons. Lahaye (1928) pointed out the similarity between thrush and pox in the pigeon. He demonstrated pox virus in many cases suspected of being thrush.

**Diagnosis.** Observation of the characteristic proliferative, relatively non-inflammatory lesions, together with resultant heavy growth on primary cultures, serve to diagnose thrush. Because of the possibility of cultivation of *M. albicans* from apparently normal tissues, an original heavy growth is considered essential for diagnosis. The recognition of spores and more especially hyphae in fresh smear preparations is attended with some difficulty.

**Course.** Young birds are more susceptible to mycosis of the digestive tract than are older birds. Thus as an infected group of birds grow older they tend to overcome the infection. Jungherr (1933a) observed an outbreak in which the losses amounted to 10,000 chicks out of 50,000 that were less than 60 days of age. He also reported (1934) that turkeys under four weeks of age succumbed rapidly to infection, but that outbreaks in birds three months of age resulted in a high percentage of recoveries.

**Prophylaxis and treatment.** Since mycosis of the digestive tract is apt to be related to unhygienic, insanitary, overcrowded conditions, these factors should not be allowed to exist, or should be corrected. Jungherr (1933b) found that denatured alcohol and coal-tar derivatives were ineffective as disinfectants and suggested that iodine preparations be used. As a treatment he recommends that following an Epsom salt flush, one level teaspoonful of powdered blue stone (copper sulfate) be added to each 2 gallons of drinking water in nonmetal containers every other day during one week. Hinshaw recommends that a 1:2,000 solution of copper sulfate for turkeys be used as the sole source of drinking water during the course of the outbreak. Affected birds should be segregated. Lesions in the mouth can be treated by local application of a suitable antiseptic. The appearance of the disease in very young chicks suggests the surface of the egg as a source of infection. Such a possibility could be removed by dipping the eggs in an iodine preparation prior to incubation.

## REFERENCES

Benham, R. W.: 1931. Certain Monilias parasitic on man. Jour. Infect. Dis. 49:183.

Eberth, J.: 1858. Einige Beobachtungen von pflanzlichen Parasiten bei Thieren. Arch. f. path. Anat. u. Physiol. 13:522.

Hinshaw, W. R.: 1933. Moniliasis (thrush) in turkeys and chickens. Fifth World's Poultry Cong., Paper 97, 190.

Jungherr, E.: 1933a. Observations on a severe outbreak of mycosis in chicks. Jour. Agr. Res. 46:169.

————: 1933b. Studies of yeast-like fungi from gallinaceous birds. Storrs Agr. Exper. Sta., Bul. 188.

————: 1934. Mycosis in fowl caused by yeast-like fungi. Jour. Am. Vet. Med. Assn. 81:500.

Klee, R.: 1899. Vergiftungen bei Geflügel. Jahresber. Vet. Med. 19:236.

Lahaye, J.: 1928. Maladies des pigeons et des poules, des oiseaux de bassecour et de volière. Remouchamps: Steinmetz-Haenen. 393.

Martin, D. S., Jones, C. P., Yao, K. F., and Lee, Jr., L. E.: 1937. A practical classification of the Monilias. Jour. Bact. 34:99.

Schlegel, M.: 1912. Soorkrankheit bei Hühnern. München. tierärztl. Wochenschr. 56:63.

————: 1921. VII. Soorkrankheit bei Gänsen. Zeitschr. f. Infektionskr. der Haustiere 21:204.

Stovall, W. D.: 1939. Classification and pathogenicity of species of Monilia. 202. Third Internat. Cong. for Microbiol., New York.

Worley, G., and Stovall, W. D.: 1937. A study of milk coagulation by Monilia species. Jour. Infect. Dis. 61:134.

Zürn, F. A.: 1882. "Durch Schimmelpilze hervorgerufene Krankheiten des Geflügels." Die Krankheiten des Hausgeflügels, Weimar. 129–35.

## "SARCOSPORIDIOSIS"

Sarcosporidiosis appears to be neither widespread nor economically important in birds, at least in the United States. The striated muscles of mammals are involved principally, although reptiles and birds are sometimes affected. It is especially common in sheep, swine, cattle, and horses. A few cases in man have been reported.

**Occurrence.** Erickson (1940) listed eight orders, thirteen families, nineteen genera, and twenty species of birds as being affected. Some of the better known hosts are the chicken, domestic mallard, wild mallard, black duck, gadwall, American pintail, blue winged teal, shoveller, turkey vulture, and English sparrow. Ducks are especially likely to be affected, and Erickson states that all recorded cases are in puddle or dabbling varieties. Hall (1925) in listing a case in a domesticated duck from a market in Washington, D. C., suggested that the infection might be of economic importance because the flesh would have been judged unfit for consumption, but there seem to be no other similar reports. Beaudette (1941) suggests that sarcosporidiosis is probably widespread but escapes notice because of the absence of symptoms in affected birds, and infection is not discovered unless the muscles are exposed. Reports of infection in chickens are uncommon, and include Germany, Kühn (1865); the United States, Stiles (1894) and Hawkins (1943); Hungary, von Ratz (1908); Bulgaria, Krause and Goranoff (1933); and Brazil, Reis and Nobrega (1936).

**Symptoms, lesions, diagnosis.** The lesions may be so small as to escape detection except by microscopic examination. When larger and present in

considerable numbers, the musculature has a finely streaked or "wormy" appearance (Fig. 17.6). The individual sarcocysts, frequently called Miescher's tubes or sacs, are usually elongated masses, the long axis of which are parallel to the muscle fiber. Large cysts are sometimes referred to as Balbiania. The sarcocysts in the case reported by Stiles (1893) were 1.0 to 6.0 mm. in length by 0.48 in breadth; in the case reported by Mathews (1930), they were 1 × 3 mm. These are larger than most cases reported in birds, but some reports in mammals range up to 5 cm. in length. An individual cyst when removed has a whitish or creamy appearance and is cylindrical with somewhat pointed ends and appears slightly lobulated on the surface. The cyst is divided by septa into compartments. The compartments in a mature cyst are filled with spores (Rainey's corpuscles) which are variously described as banana, crescent, sickle, or comma shaped. The spores are 3 to 15μ in length and 1 to 4μ in width. The compartments in the center of an old cyst tend to undergo degeneration. Mathews (1930) called attention to a variation in connective tissue and inflammatory response which was proportionately greater around cysts with more degeneration (Fig. 17.7). The sarcocysts start development within muscle fibers, but as they enlarge the fibers are destroyed and the larger



Fig. 17.6. Severe sarcosporidiosis in a duck. (Becker, Iowa State College.)

cysts are intermuscular. Apparently the sarcocysts do not seriously injure their hosts. Most reports are on birds which were considered normal when killed. Very heavy infestations may possibly cause symptoms. Mice may be killed by heavy doses, and this suggests that the same may be true in larger animals and birds.

**Etiology.** Following the discovery by Miescher (1843) of Sarcosporidia in the muscle tissue of a mouse, they were first named *Synchytrium miescherianum* by Kühn (1865). This genus was, however, already in use to describe a group of fungus-like organisms, and the genus Sarcocystis was established

by Lankester in 1882. There are names of many species in the literature designated principally with respect to the animals infected, but the organisms have not always been host-specific, and the morphological differences are not substantial except in relation to size of the cysts. Alexeieff (1913) concluded that there was no reliable ground for distinguishing the supposed species of Sarcocystis and that all belong to one species, *S. miescheriana*. Hagan (1943) suggests that *S. miescheriana* has priority if all are considered as a single species, otherwise this term would apply only to infection in the
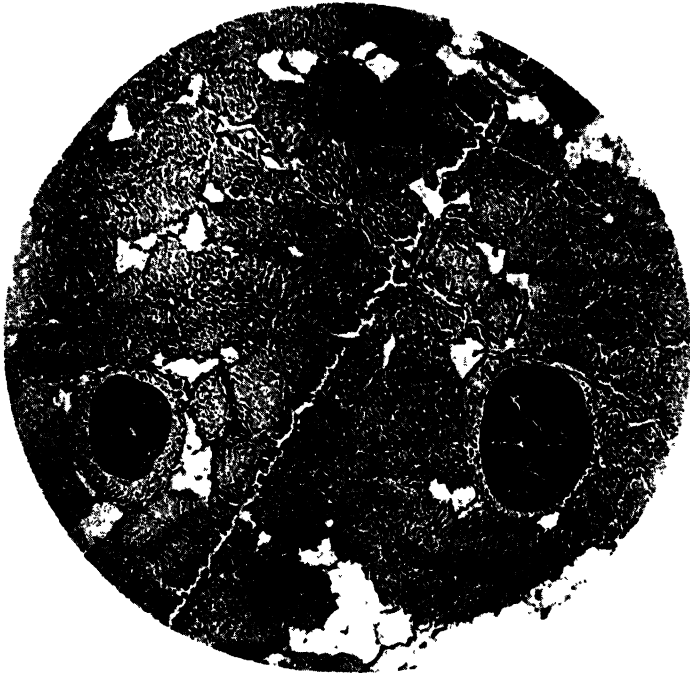


FIG. 17.7. Sarcosporidia in breast muscle of chicken. ×150. (Biester, Iowa State College.)

pig. *Sarcocystis rileyi*, Stiles (1893), has been the term applied to the presumed infective protozoan in ducks since Riley told Stiles (1893) that the lesions noted by Walsh and Riley (1869) and believed by them to be similar to *Cysticercus cellulosae* of pork were identical with Stiles' Sarcocystis. Hawkins (1943) compared sarcosporidiosis in the chicken, the mallard, the domestic mallard, and the black duck and stated that all were apparently the same species.

The life history of Sarcosporidia is incompletely known and only brief mention is made here. Although Theobald Smith (1901, 1905) and others have reported transmission particularly in mice by feeding flesh containing mature sarcocysts, it is probable that this would not have been accomplished had the eating of feces been prevented. Spindler, Zimmerman, and Jaquette
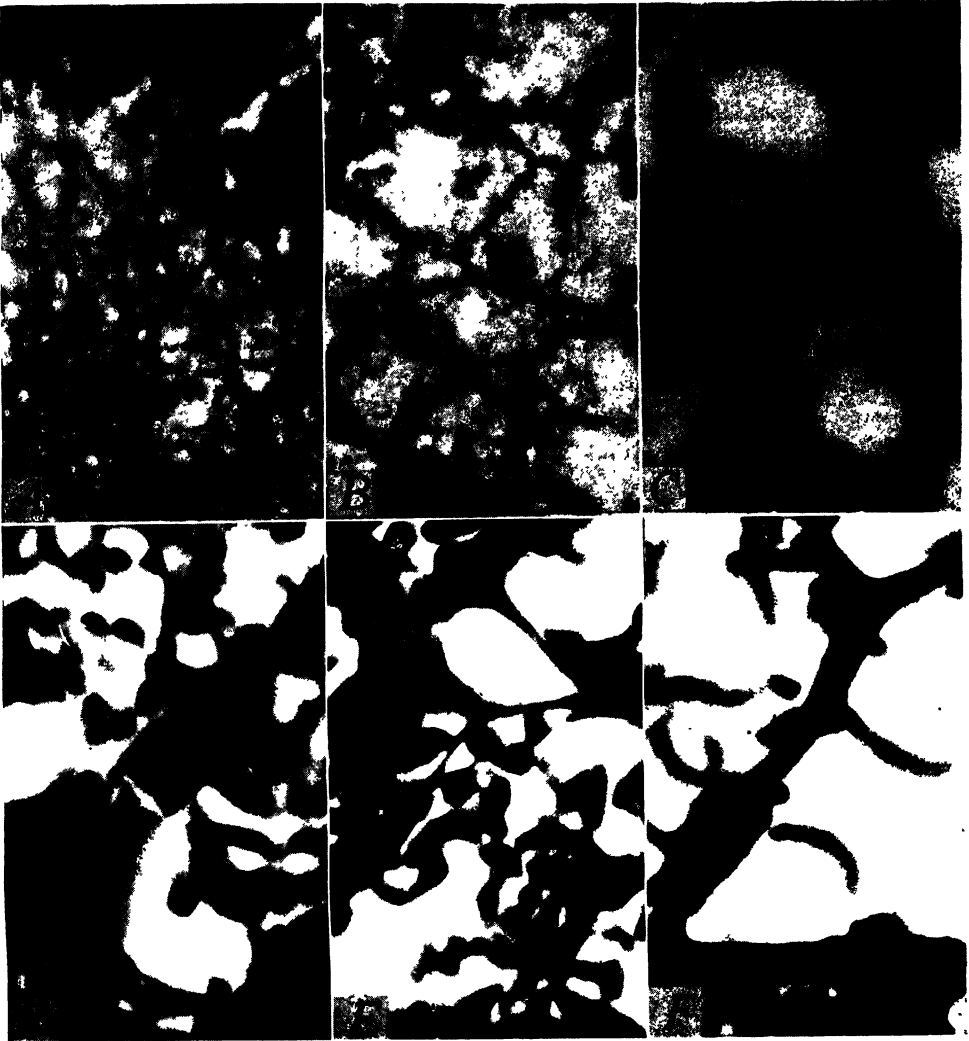
FIG. 17.8. Miescher's sacs, showing arrangement of "septa" (A, B, and C); their jointed structure (C, D, and F); and attachment of the Rainey's corpuscles (spores) to the "septa" (E and F). Sections were stained with Gram's stain. A, B, and C are from a naturally infected wild duck; D, E, and F from a naturally infected sheep. (Spindler, Proc. Helminth. Soc. Wash. 14:28, 1947.)

(1946) established infection in swine by feeding feces and/or urine from animals and birds which had been fed muscles containing sarcocysts. Flesh from infected swine was fed to pigs, dogs, cats, rats, mice, and chickens. The feces and/or urine from these animals and birds were not infective for 15 days after consumption of the infected flesh, but contained a stage of Sarcocystis thereafter which was infective for swine. These findings are in harmony with those of Negre from 1910 to 1918 (cited by Spindler *et al.,*

1946). Scott's (1930, 1943) reports which include extensive surveys of the literature and Babudieri (1932) should be consulted by anyone involved in research on sarcosporidiosis. There is a latent period of at least six weeks between the time of exposure and the development of sarcocysts in the muscles. What happens during this time or the mode of escape of infective material from infected muscle is uncertain, but presumably those processes take place through the blood stream.

There has been considerable discussion as to whether the etiological agent is a protozoan or a fungus, and Wenyon (1926) suggested that the Sarcosporidia probably are fungi. Spindler and Zimmerman (1945) reported an investigation which showed that the infective agent in swine is a fungus and not a protozoan. An Aspergillus sp. was recovered by aseptically rupturing sarcocysts or Miescher's sacs into dextrose culture solution. Young pigs injected with or fed conidia harvested from the cultures harbored typical sarcocysts in the muscles at necropsy, four to six months after exposure. A fungus like that injected was recovered on cultures from the mature sarcocysts. Spindler (1947) prepared histological sections from a sheep and a duck in a study of the internal structure of Miescher's sacs. The sacs contain a network of jointed hypha-like structures (Fig. 17.8A, B, C, and D). The septa divide the sac into compartments (Fig. 17.8B and C). These structures appear jointed (Fig. 17.8C, D, and F). Rainey's corpuscles (spores) are shown attached to the septa (Fig. 17.8E and F). The staining reaction of the structures was found to be characteristic of fungi. This was confirmed by finding a delicate septate mycelium by heating Miescher's sacs from sheep, cattle, and birds in 30 per cent KOH solution and staining the residue with lacto-phenol-cotton blue solution.

These recent findings in sarcosporidiosis will tend to redirect investigations and may hasten the procurement of definite information on many points which are not understood at present. Investigations are likely to be more intensive in mammals, particularly swine and sheep, than in birds because of the relative economic importance.

## REFERENCES

Alexeieff, A.: 1913. Recherches sur les Sarcosporidies. 1. Étude morphologique. Arch. Zool. Exp., LI, 521. (Citation from Wenyon.)

Babudieri, B.: 1932. I Sarcosporidi e le Sarcosporidiosi. Arch. f. Protistk. 76:421.

Beaudette, F. R.: 1941. Sarcosporidiosis in a black duck. Jour. Am. Vet. Med. Assn. 99:52.

Erickson, A. B.: 1940. Sarcocystis in birds. The Auk 57:514.

Hagan, W. A.: 1943. The Infectious Diseases of Domestic Animals. Comstock Publishing Co., Ithaca, N. Y. P. 489.

Hall, M. C.: 1925. Sarcocystis rileyi from the domesticated duck. Jour. Parasit. 11:217.

Hawkins, P. A.: 1943. Sarcocystis rileyi (Stiles, 1893) in the domestic fowl, Gallus gallus. Jour. Parasit. 29:300.

Krause, C., and Goranoff, S. A.: 1933. Ueber Sarkosporidiosis bei Huhn und Wildente. Zeitschr. Infektionskr. Parasit. Krank. und Hyg. Haustiere 43:261.

Kühn, J.: 1865. Untersuchungen über die Trichinenkrankheit der Schweine. Inst. d. Univ. Halle 68. (Citation from Erickson.)

Mathews, F. P.: 1930. Sarcosporidiosis in a duck. Jour. Am. Vet. Med. Assn. 76:705.

Miescher, F.: 1843. Ueber eigenthümliche Schläuche in den Muskeln einer Hausmaus. Ber. ü. d. Verhandl. Naturf. Ges. Basel, V, 198. (Citation from Wenyon.)

Reis, J., and Nobrega, P.: 1936. Tratado de doencas das aves. São Paulo, Brazil.

Scott, J. W.: 1930. The Sarcosporidia. A critical review. Jour. Parasit. 16:111.

————: 1913. Life history of Sarcosporidia, with particular reference to *Sarcocystis tenella*. Wyo. Agr. Exper. Sta., Bul. 259:1.

Smith, T.: 1901. The production of sarcosporidiosis in the mouse by feeding infected muscular tissue. Jour. Exper. Med. 6:1.

————: 1905. Further observations on the transmission of *Sarcocystis muris* by feeding. Jour. Med. Res. 13:129.

Spindler, L. A.: 1947. A note on the fungoid nature of certain internal structures of Miescher's sacs (Sarcocystis) from a naturally infected sheep and a naturally infected duck. Proceed. Helminth. Soc. of Wash. 11:28.

———— and Zimmerman, Jr., H. E.: 1945. The biological status of *Sarcocystis*, Jour. Parasit. 31 (Dec. suppl.) :13.

————, Zimmerman, Jr., H. E., and Jaquette, D S.: 1946. Transmission of *Sarcocystis* to swine. Proc. Helminth. Soc. of Wash. 13:1.

Stiles, C. W.: 1893. Notes on parasites—18: On the presence of sarcosporidia in birds. Bur. An. Ind., U.S.D.A., Bul. 3:79.

————: 1894. Notes sur les parasites. Bul. Soc. Zool., France. 19:160.

Walsh, B. D., and Riley, C. V.: 1869. A measly wild duck. Am. Entomol. 1:89.

Wenyon, C. M.: 1926. Protozoology. Ballière, Tindall, and Cox, London.

von Ratz, I.: 1908. Az izmokban élosködö véglényekröl es a Magyar fauna ban elöforduló fajaikród. Allattani Közlemények 7:177. (Citation from Erickson.)

CHAPTER EIGHTEEN

# THE AVIAN LEUKOSIS COMPLEX

By ERWIN JUNGHERR, *Department of Animal Diseases,*
*University of Connecticut, Storrs, Connecticut*

★   ★   ★

## INTRODUCTION

Leukosis signifies a group of diseases which is characterized by autonomous proliferation of the precursors of leukocytes. The term has largely replaced the older name leukemia (white bloodedness) principally because changes in the circulating blood, as implied by the name, are not an invariable pathologic feature. For the forms without blood involvement qualifying terms such as "aleukemic" and "pseudoleukemic" have been suggested.

Avian leukosis was first studied in a systematic way by Ellermann, and fowl paralysis by Marek, at the beginning of the present century. At first held to be completely separate, both of these conditions were shown during the last decade to have a similar epidemiology, to share a tendency toward tumor formation and, in some intances, to have an etiologic relationship. Most of the studies have dealt with the common fowl. The knowledge on corresponding diseases in other species of birds is incomplete. However, certain species of birds have proved susceptible to experimental transmission of chicken leukosis, and reciprocal transmission of fowl paralysis from chicken to pheasant (and perhaps turkey) seems probable.

While taxonomically the leukoses cannot be separated from avian neoplasia, the common features within and the economic importance of the leukosis group provide theoretical and practical reasons for bringing the available knowledge together under one heading. It is, therefore, proposed to discuss fowl paralysis, leukosis, and kindred conditions under one chapter entitled "The Avian Leukosis Complex" without prejudicing future etiologic classification of the various forms.

The present development of our knowledge on the avian leukosis complex is outlined briefly in the historical part; the various pathologic manifestations are discussed as independent entities similar to the way they present themselves in practice, while cause and control are taken up from a common point of view.

## HISTORICAL

The varied opinions on, and the practical importance of, the avian leukosis complex are reflected in the large number of references to this disease

group in the literature. Comprehensive reviews have been prepared by
Biely and Palmer (1932), Jármai (1934), Olson (1940), and recently by
Engelbreth-Holm (1942), and Furth (1946). With due apology to authors
not mentioned here, the literature is selected from the standpoint of tracing
the contributions which form the framework of our present concept of the
avian leukosis complex.

**Fowl paralysis.** Under the term polyneuritis, Marek (1907) described
a disease of chickens which was characterized by lameness and pathologic
enlargement due to mononuclear infiltration of the peripheral nerves. He
differentiated it from Eijkman's beriberi-like polyneuritis in chickens now
known to be caused by thiamin ($B_1$) deficiency. In studying a similar disease
in the North Atlantic States, Kaupp (1921) observed its frequent association
with blindness. The first positive transmission experiments were reported
by Van der Walle and Winkler-Junius in Holland (1924). The disease was
studied by Doyle (1926, 1928), from a pathologic point of view, and by
Pappenheimer and his associates (1926, 1929a, 1929b), who introduced the
term *neurolymphomatosis gallinarum*. The last-mentioned authors pointed
out the frequent association of visceral lymphomata originating from the
ovary with infiltrative lesions in peripheral nerves, brain, and iris, and pro-
duced evidence of the transmissibility of the disease in about 25 per cent of
the experimental birds. They believed neurolymphomatosis to bear no
relationship to Ellermann's form of leukosis. The transmissibility of neuro-
lymphomatosis has been questioned frequently in the literature (Olson,
1937).

In a differential study of neurolymphomatosis and the lymphatic form of
leukosis, Furth (1935) pointed out that the former is of frequent occurrence,
associated with clinical paresis, and characterized by small-cell lymphocytic
infiltration of the peripheral nerves and viscera, without blood or bone
marrow involvement; the condition was found by him to be transmissible
only by viable cells.

**Fowl leukosis.** Leukosis in the common fowl was first recorded by
Caparini in 1896, according to Olson (1940). Extensive experimental
studies of the condition were undertaken by Ellermann and his associates
(1922, 1923), who recognized three general forms of avian leukosis, namely
leukemic or aleukemic myeloid leukosis, intravascular "lymphoid." i.e.,
erythroid leukosis, and extravascular lymphatic leukosis, all of which were
considered to be transmissible and caused by the same filtrable virus.

This subject presented an intriguing problem to modern leukemia re-
search in man and animals and was reopened in this country by Furth
(1931a), who discovered a new strain of readily transmissible leukosis which
conformed to Ellermann's intravascular "lymphoid" form; a rare subvariety
with little blood involvement was described as anemic erythroleukosis

(1931b). The characterization of erythroleukosis was soon confirmed and extended, especially in Denmark by Engelbreth-Holm and Rothe Meyer (1932), who suggested the term erythroblastosis, and by Oberling and Guérin (1934) in France.

**Granuloblastosis.** Leukemic myeloid leukosis of Ellermann was observed by Furth (1931a) and others in serial transmission experiments with strains of erythroleukosis. "Pure" strains of erythroleukosis and granuloblastosis were observed by Jármai (1930) and Nyfeld (1934), respectively. Although there are pathologic and especially hematologic differences between erythro- and myeloid leukosis (relative preponderance of erythroid or myeloid precursors in the blood), the two tend to occur in a "mixed" form (Furth, 1931a) and are conceded to be caused by the same filtrable agent. Olson (1936) discussed this disease under the heading of granuloblastic leukosis.

**Lymphomatosis.** Lymphatic leukosis represented the only extravascular form in Ellermann's classification of transmissible avian leukosis. Later his erstwhile collaborators, Andersen and Bang (1928), expressed doubt as to its transmissibility. That this disease constituted an independent nontransmissible entity was maintained by Mathews and Walkey (1929), who suggested the designation "lymphadenoma" and separated it sharply from neurolymphomatosis. This view was upheld by Feldman (1932), and by Feldman and Olson (1933), who used the term "lymphocytoma" for the aleukemic neoplastic disease for which transmissibility had not been demonstrated and the type cell of which was the undifferentiated lymphocyte. Fenstermacher (1932, 1934, 1936) reported extended observations on the non-transmissibility of lymphocytoma and its familial incidence. Attempts by Olson and Zeissig (1936) to distinguish lymphocytoma from neurolymphomatosis by serologic means were inconclusive. Oberling and Guérin (1934) recognized differences between the nontransmissible extravascular forms and the transmissible intravascular ones. Furth (1935) agreed with the definition of lymphocytoma insofar as the "spontaneous" extraordinary enlargements of the liver were concerned, which condition he termed "hepato-lymphomatosis."

While it would appear from the foregoing statements that three separate entities have to be recognized, namely, neurolymphomatosis, transmissible erythro-myeloleukosis, and nontransmissible lymphocytoma, other studies tended to break down the sharp boundaries. Johnson (1932) was unable to differentiate lymphocytoma from the visceral lymphomata which had been described by Pappenheimer et al. (1926) in cases of neurolymphomatosis, and believed this association to be so common that he proposed the generic term "lymphomatosis" for the specific "neural" and "visceral" subdivisions. Furth (1933) developed a transmissible agent (Strain 2) which was capable of causing what he also termed lymphomatosis and at times myelocytomatosis

and endothelioma. In distinction from neurolymphomatosis, he (1935) considered lymphomatosis to be a rare disease which was not associated with clinical paresis; pathologically it was characterized by anemia, large-cell lymphocytic leukemia, and tumorous infiltrations of the same cell-type in the visceral organs and occasionally the peripheral nerves; the disease proved easily transmissible by cell-free material. Since the term lymphomatosis was thus used in an etiologically and pathologically equivocal sense by Johnson and Furth, Jungherr (1937) studied field cases of neurolymphomatosis and lymphocytoma-like conditions in transmission experiments; the results indicated that the pathologic range of such transmissible strains may include both types of lymphomatosis, and favored the Johnson conception of lymphomatosis. Recent experimental studies on the transmissibility of visceral lymphomatosis by Davis and Doyle (1947) indicated, however, that the incidence of the visceral type alone could be increased by inoculation. Supported by the analysis of extensive field material this group of authors (Davis, Doyle, Walkey, and Cenker, 1947) considered visceral lymphomatosis as a separate entity, distinct from neural lymphomatosis.

As stated before, endotheliomata were observed by Furth (1933) in passages of Strain 2. Starting with material from a case of neurolymphomatosis, Jungherr (1937) likewise observed endotheliomata. It would appear that this condition is sometimes an expression of lymphomatosis.

An uncommon hypertrophic osteopathy of chickens has been described in general ornithopathology under various terms such as hyperplastic ostitis (Reinhardt, 1930) and diffuse osteoperiostitis (Pugh, 1927). Transmission studies on this condition by Jungherr (1935) and Jungherr and Landauer (1938) tended to show that certain strains of lymphomatosis carry hypertrophic-osteopathic potentialities, for which the term "osteopetrosis gallinarum" was suggested.

**Lymphoid tumors.** Olson (1941) described a transplantable lymphoid tumor of the chicken with an unusually short incubation period that, according to gross and histopathologic features, would fall under the classification of lymphomatosis. Designating such a tumor as transplantable lymphosarcoma, Pentimalli (1941) made similar observations.

While at first, the relationship of transplantable lymphoid tumors to the avian leukosis complex was not clear, recent intensive studies of the Olson tumor and similar highly malignant tumor strains by Burmester and his associates hold promise of clarifying the etiologic relationship of the various forms within the avian leukosis complex. In their hands (Burmester, Brandly, and Prickett, 1944) the Olson tumor maintained its virulence almost unabated when frozen slowly and stored at $-65°$ to $-76°$ C. for 391 days. By intraperitoneal injections of young chickens with tumor tissues which morphologically were indistinguishable from lymphocytoma, visceral

lymphomatosis, or "lymphoid tumors," Burmester and Prickett (1945) were able to develop several additional highly malignant tumor strains. The donors were obtained from a flock which had been kept for genetic studies in isolation for five years and was affected, aside from sporadic cases of coccidiosis, with no other known disease except naturally occurring avian lymphomatosis. In subsequent studies such transplantable tumor tissues and the plasma of lymphoid tumor-bearing chickens were shown to contain a filtrable agent, capable of inducing visceral lymphomatosis and osteopetrosis after an incubation period from two to six months (Burmester, Prickett, and Belding, 1946a). The filtrable agent proved to be sedimentable to considerable extent by high-speed centrifugation. In that, the agent of lymphomatosis tended to occur in lower concentration in the sediment and to be endowed with a longer incubation period, than that of osteopetrosis, thereby suggesting two etiologically different agents (Burmester, 1947a). The existence of filtrable agents producing lymphoid tumors and osteopetrosis was confirmed by serial passage in chickens (Burmester and Cottral, 1947). The controversial question of the transmissibility of avian visceral lymphomatosis was restudied. Although finding variations in the transmissibility of the disease from different donors (Burmester and Dennington, 1947), avian lymphomatosis could be propagated with cellular as well as cell-free preparations (Burmester, 1947b). In this series the incidence of osteopetrosis and neural lymphomatosis was surprisingly low which suggested different etiologic agents for the latter conditions. In consequence of these observations Burmester (1947c) expressed the opinion that many cases of natural lymphomatosis carry masked lymphoid tumor agents.

In immunologic studies of lymphoid tumors, Burmester and Prickett (1944) found large doses of viable tumor (Olson) cells to be fatal, but small doses which permitted regression of the take, to induce a lasting solid immunity. Olson (1945a, b) pointed out that the immunizing capacity could be enhanced by serial passage and depended on the viability of the implants although it persisted to some extent after reducing growth-activity by freezing or heating. Normal or embryonic tissues lacked such immunogenic properties (Olson, 1947).

Chickens immunized against a transplantable lymphoid tumor were no more resistant to spontaneous neural or visceral lymphomatosis than comparable controls in the experience of Burmester, Prickett, and Belding (1946b), and of Olson (1947). The latter author found also chickens spontaneously affected with neural lymphomatosis to be fully susceptible to implants of a lymphoid tumor.

**Myelocytomatosis.** Aleukemic myeloid leukosis was considered by Ellermann (1923) as a subvariety of transmissible myeloid leukosis. The disease was ordinarily associated with tumor formation. Pentimalli (1915) appar-

ently first described a spontaneous chicken tumor which was composed almost exclusively of myelocytes with the characteristic granulations of the mature eosinophil and heterophil. Mathews (1929) believing the tumor to be analogous to chloroma in man, suggested the term "leukochloroma." He failed to show transmissibility of the condition. Feldman (1932) regarded it as a definite neoplastic process which he classified as myelocytoma. In a passage experiment with lymphomatosis Strain 2, Furth (1933) observed cases of myelocytoma with myelocytic blood involvement (myelomatosis). This apparently constitutes one of the few recorded instances of transmissible myelocytomatosis.

**The unitarian view.** In general the extended investigations of the avian leukoses by Furth tended to show that each transmissible strain represents an etiologic unit because of its more or less definite, if occasionally wide [e.g., Strain 2 (1933)] pathologic range which can be established only by observing the behavior of the strain through several passages. Quite in contrast to this concept is the unitarian point of view which assumes that a single etiologic agent is responsible for all of the various pathologic manifestations of the avian leukosis complex. On the basis of transmission experiments with neurolymphomatosis and fowl leukosis, Patterson and co-workers (1932, 1934, 1936) concluded that "fowl leukosis" can be subdivided into erythroid, myeloid, lymphoid (including lymphocytoma), nerve, eye, and mixed types, all of which were considered to be different expressions of the same transmissible disease. Lee (1942) reported that serums of ducks, turkeys, and chickens previously immunized with material from myeloid or lymphoid leukosis possessed neutralizing properties for both the homologous and the heterologous variety of the leukosis agent.

Johnson (1934), in continuation of his work on lymphomatosis (1932), considered erythro- and myeloleukosis likewise to be due to the causal agent of lymphomatosis, and suggested the inclusive term "hemocytoblastosis" for the various morphologic types. This term was based upon the concept of Jordan and Johnson (1935) and Jordan (1936) that the hemocytoblast of the marrow stroma is the primitive reticulum stem cell which gives rise to both the erythroblastic and granuloblastic series. This was in conformity with Ringoen (1934), and in contrast to the theory of attributing erythrogenesis and granulocytogenesis to the endothelial cells of the venous sinuses, advanced by Doan, Cunningham, and Sabin (1925). The unitarian view was further supported by filtration studies through collodion membranes by Johnson and Bell (1936), who were thereby unable to separate the agents of leukosis and lymphomatosis, and by the recent studies of Johnson (1940). Hall, Bean, and Pollard (1941) developed a strain of hemocytoblastosis from original neurolymphomatosis material, and thus amplified the evidence in favor of a unitarian view. The strain was carried by intravenous injection

of chicken embryos through thirty passages and produced nearly 41 per cent takes in embryos or one-week-old chicks (Hall and Pollard, 1943).

Emmel (1939) advanced a similar unitarian view and likewise used the inclusive term "hemocytoblastosis." He believed, however, that the disease was incited by a variety of etiologically unrelated stimuli such as Salmonella toxicoses, anoxemias, and avitaminoses. Hemocytoblastosis was defined by him as an increase or decrease in leukocytes accompanied by the appearance of immature and degenerative blood cells in the circulating blood. Blount (1939) has shown that similar blood pictures occur in physiologic transition stages from embryonic to adult life and in various unrelated diseases; he considered hemocytoblastosis to be a certain "type of myeloid response, not a disease *per se*" and nonessential in the development of fowl paralysis.

**Relation of leukosis to virus tumors.** Until recently leukosis was thought to be unrelated to transmissible sarcomas and similar tumors of the fowl which have been studied extensively by Rous, his associates, and other workers (for ref. see Claude and Murphy, 1933; Foulds, 1934). Oberling and Guérin (1933a, b), however, obtained new evidence on the production of malignant tumors of the Rous type with the virus of transmissible leukosis, and a similar polyvalent strain was studied by Rothe Meyer and Engelbreth-Holm (1933) and by Engelbreth-Holm (1935). Troisier (1935) was inclined to assume the unity of the leukosis and sarcoma virus. A strain of leukosis described by Jármai (1935) produced fibrosarcomas at the point of injection in leukosis-refractory birds. These various studies suggested that certain leukosis strains produced the typical homologous disease on intravenous administration, but tumors at the site of injection, especially if the inoculum consisted of tissue material rather than blood.

These observations stimulated a large amount of investigational work on the relation of leukosis to sarcoma, the results of which seemed to indicate that simple, mixed, and complex strains occur (Furth, 1936a).

Simple or pure strains maintain their pathologic identity in successive passages; that is, they reproduce the disease of the original donor. They are exemplified by the Rous sarcoma or erythroleukosis (Strain 1) of Furth (1931a), which Stubbs (1938) tested for tumor-producing properties over several years, with negative results. A recently isolated Canadian strain of erythroleukosis likewise failed to produce neoplasm at the site of inoculation (Wickware, 1943, 1946).

If leukosis and sarcoma-like processes occur in the same donor and prove to be dissociable in subpassages, the conception of "mixed" strain is applicable. Furth (1936b) observed an osteochondrosarcoma (Strain 12) in a bird inoculated with lymphomatosis (Strain 2) (1933) and showed that in successive transplantations both pathologic components occurred either alone or in combination. However, later culture studies of the virus in vitro

by Furth and Breedis (1937) suggested that it may have been a complex strain. From a spontaneous ovarian tumor which had both lymphomatous and sarcomatous characters, Jungherr (1937) developed agents of lymphomatosis and sarcomatosis, the latter of which was carried as such through many subpassages by Cole (1941).

Complex strains apparently are due to a single agent which can stimulate both primitive blood cells and fibroblastic cells. Stubbs and Furth (1935) described the interesting Strain 13 which produced sarcoma on subcutaneous or intramuscular inoculation, and diffuse endothelial sarcomatosis in the blood-forming organs associated with erythroleukosis, when injected intravenously. The strains studied by Oberling (1933a, b), Rothe Meyer (1933), and Jármai (1935) may have been of a similar order (Furth, 1936a).

**Discussion of literature.** A cursory review of the literature on the avian leukosis complex emphasizes primarily the complexity of the problem. The boundaries of the disease group, due to the apparent existence of complex leukosis-sarcoma strains, are distinct. *If pressed for a definition one might venture to say that the diseases of the avian leukosis complex are such which are primarily characterized by autonomous proliferation of essential blood-forming cells, and are as a rule, due to oncogenic viruses.* An etiologic classification, desirable as it may be, is not possible in the present state of our knowledge. The focal point in question is the etiologic unity or disunity of fowl paralysis, leukosis, and certain neoplasms. Furth's conception of the essential pathologic and etiologic specificity of various oncogenic viruses sounded an important warning against sweeping generalizations.

Significant confirmation has come for this view from the work of Davis and Doyle and particularly from that of Burmester and his associates. Both groups of workers considered neural and visceral lymphomatosis to represent distinct entities. The extensive studies at the United States Regional Poultry Laboratory by Burmester and his co-workers indicated the relative frequent occurrence of transplantable lymphoid tumors and their association with filtrable agents capable of inducing visceral lymphomatosis with or without osteopetrosis. Thereby, the concept of "lymphocytoma" as a non-transmissible form of lymphomatosis has become untenable. The trend of modern research is definitely in the direction of etiologic subdivision of the various forms of the avian leukosis complex.

Considerable agreement exists on the morphology of the various expressions many of which can be diagnosed with reasonable certainty by the unaided eye. Even the most detailed histologic examination is incapable of indicating transmissibility or pathologic potentiality of a given case. Many autopsy records on poultry testify to the economic importance of the avian leukosis complex, far in excess of other neoplastic or infectious diseases of adult birds. Thereby the problem of classification is distinctly removed from

the academic sphere and assumes a major practical aspect. Descriptions of strains and strain numbers can serve only as prototypes, not as a basis of classification. Under the existing multiplicity of and confusion in terms, ornithopathology is poorly equipped to guide the poultry industry in any contemplated control program.

Realizing this situation, a committee of pathologists (Jungherr, Durant, and Lee, 1941) cooperating with the United States Regional Poultry Research Laboratory at East Lansing, Michigan, has suggested a tentative pathologic classification, which is used here.

### TENTATIVE NOMENCLATURE OF THE AVIAN LEUKOSIS COMPLEX[1]

It is understood that these various forms can be leukemic, subleukemic (less than 100,000 leukocytes per mm.$^3$ of blood) (Furth, 1933), or aleukemic.

Lymphomatosis (Johnson, 1932)
    neural (Pappenheimer et al., 1926)
    ocular (Pappenheimer et al., 1926)
    visceral (Johnson, 1932)
    osteopetrotic (Jungherr and Landauer, 1938)
Erythroblastosis (Furth, 1931a; Engelbreth-Holm and Rothe Meyer, 1932)
Granuloblastosis (Olson, 1936)
Myelocytomatosis (Furth, 1933; Mathews, 1929)

### OTHER TUMORS

Sarcomatosis

etc.

**Comment.** The object of the suggested classification is to facilitate uniformity in terminology and epidemiologic interpretation of data from various laboratories. The scheme does not imply classification on an etiologic basis.

With respect to particulars, the term avian leukosis complex points out the theoretical and practical necessity of looking at the problem as a whole. The concept of lymphomatosis reflects the practical observations of the co-occurrence of the various subdivisions. Present indications are, however, that the neuro-ocular, visceral, and osteopetrotic forms constitute separate entities. The terms erythroblastosis and granuloblastosis replace that of "leukosis," which had been given such a variety of interpretations in the

---

[1] Adopted by conference of investigators in this field (July 15–17, 1940, East Lansing, Michigan) and revised October 21–23, 1941. References cited give pathologic descriptions, not necessarily the names of the corresponding types.

literature that it seems inadvisable to use it in the detailed classification. Myelocytomatosis being rarely leukemic, and more commonly neoplastic, forms a link between the avian leukosis complex and neoplasia. The expression "other tumors" calls attention to the fact that the avian leukosis complex is considered a tumor of the blood-forming organs. The suggested scheme represents a working hypothesis, subject to revision on further evidence.

## NEURAL LYMPHOMATOSIS

**Synonyms.** Fowl paralysis, range paralysis, polyneuritis (Marek, 1907), neuritis (Doyle, 1926), neurolymphomatosis gallinarum (Pappenheimer et al., 1926), neurogranulomatosis (Lerche and Fritzsche, 1934).

Paretic symptoms may be observed as accompanying a variety of diseases
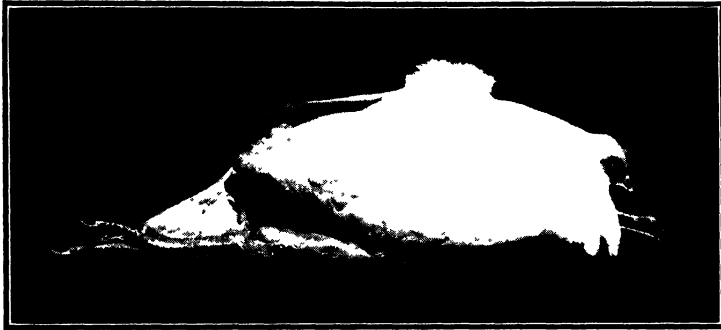


Fig. 18.1. Neural lymphomatosis. Typical clinical position in advanced case. (L. P. Doyle, Jour. A.V.M.A.)

such as tuberculosis, staphylococcosis, fowl cholera, helminthiasis, coccidiosis, botulism, avitaminosis, lead and salt poisoning, etc. To differentiate such conditions from true fowl paralysis, as discussed below, the former may be grouped under the term "symptomatic paralysis," according to Bayon (1932).

**Occurrence.** The disease has been observed in every major poultry-producing country of the world. It attacks primarily young birds between two and five months of age, but has been seen as early as three weeks of age and as late as the second year of production. All breeds and both sexes are susceptible. The losses are variously estimated as from 5 to 25 per cent in affected flocks. Under practical conditions the disease often makes its appearance when the birds are first turned out on range; then after a period of quiescence the disease apparently resumes its course in the visceral form when the birds are put into the laying houses.

**Symptoms.** The clinical signs of the disease are usually those of asymmetric progressive paresis of the leg, wing, or neck, the paresis being either spastic or flaccid in character. In the beginning the affected leg may show

inward curving of the toes, weakness, or incoordination. Later on, the bird has a tendency to hold one foot stretched forward or backward, a position which is quite characteristic (Fig. 18.1). If both legs are affected the animal moves with difficulty, in a squatting position. Involvement of a wing is indicated by drooping of the extremity (Fig. 18.2); if one tries to spread the wings the diseased one often gives the impression of increased resistance. Paresis of the neck may be suggested by low carriage of the head and incipient torticollis; affection of the deep muscles and the nerves, especially the vagus,
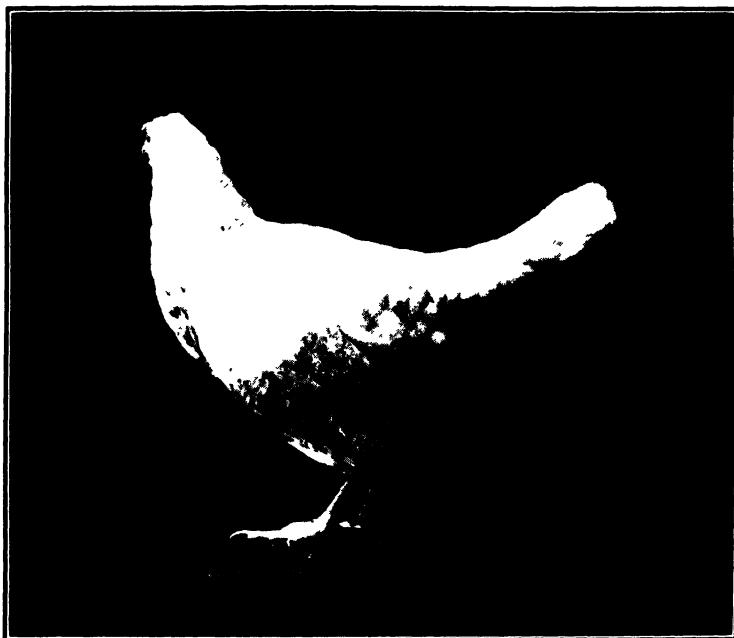


Fig. 18.2. Involvement of a wing. as indicated by drooping of extremity. (L. P. Doyle. Jour. A.V.M.A.)

may lead to dilatation of the crop and gasping symptoms. Durant and Mc-Dougle (1945) considered soiled, damp, "front" feathers under the beak and along the throat to be a symptom of neural lymphomatosis and attributed it to involvement of the posterior cranial and the anterior cervical nerves. Locomotory disturbances are often associated with systemic reactions such as loss of weight, paleness, anorexia, and diarrhea, although the appetite not infrequently remains good. The symptoms of true fowl paralysis are not specific, and vary widely in intensity.

**Pathology.** The gross anatomic features of the disease are characterized by localized or occasionally diffuse grayish, soft swellings of the peripheral nerve trunks. The femoral portion of the sciatic trunk is commonly affected (Fig. 18.3). This nerve can be observed easily by lifting up the large triangu-

lar adductor muscles on the median surface of the thigh under which its two strands run parallel with the femoral artery. The normal nerve is uniform in width, white, and cross-striated. Loss of striation is suggestive of the disease. Bilateral comparison permits the detection of mild alterations. For detailed examination the distal and proximal ramifications of the nerve should be exposed. Changes may be seen in the intrapelvic part of the trunk, the so-called sciatic plexus, which originates from four spinal roots and is located under the middle lobe of the kidney. The lumbar and celiac plexuses stand out very prominently, if affected. Some portion of the brachial plexus may be involved, although there is no constant correlation between neuro-
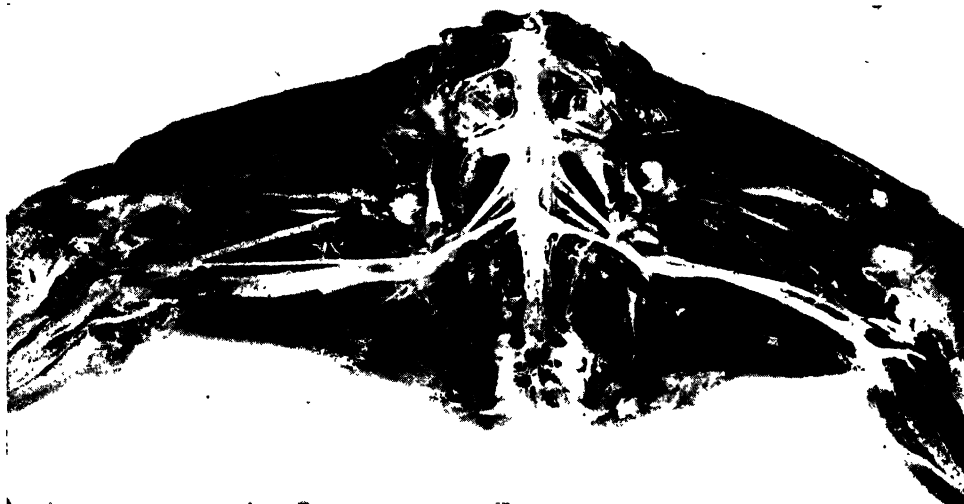


FIG. 18.3. Neurolymphomatosis. Femoral portion of right sciatic nerve thickened. (Iowa State College.)

muscular disturbances and regional nerve lesions. Vagus affection is apt to occur if respiratory symptoms are present. The dorsal ganglia of the peripheral nerves, especially in the region of the brachial plexus roots, often undergo grayish enlargement which may extend into the spinal cord and form tumor-like masses (Fig. 18.4).

Unless the pathologic form of visceral lymphomatosis is also present the visceral organs appear normal in uncomplicated cases of fowl paralysis. The spleen is usually of normal size; the thymus lobes may show glandular enlargement (Jungherr, 1933). According to Johnson (1934) the bone marrow of the ulna and radius, which ordinarily is fatty and aplastic in mature birds, may revert to the state of hyperactivity often seen in young normal birds.

New systemic lesions were described by Blakemore (1939), and Blakemore and Dalling (1939), who in transmission experiments of neuro-

lymphomatosis with liver emulsions observed whitish, ill-defined areas especially in the heart and liver substance corresponding microscopically to focal necrosis. These lesions occurred within a short time after inoculation and were followed by infiltration with various mononuclear cells. Typical nerve lesions developed in protracted cases. The authors suggested that fowl paralysis in early stages was characterized by systemic necrotizing-
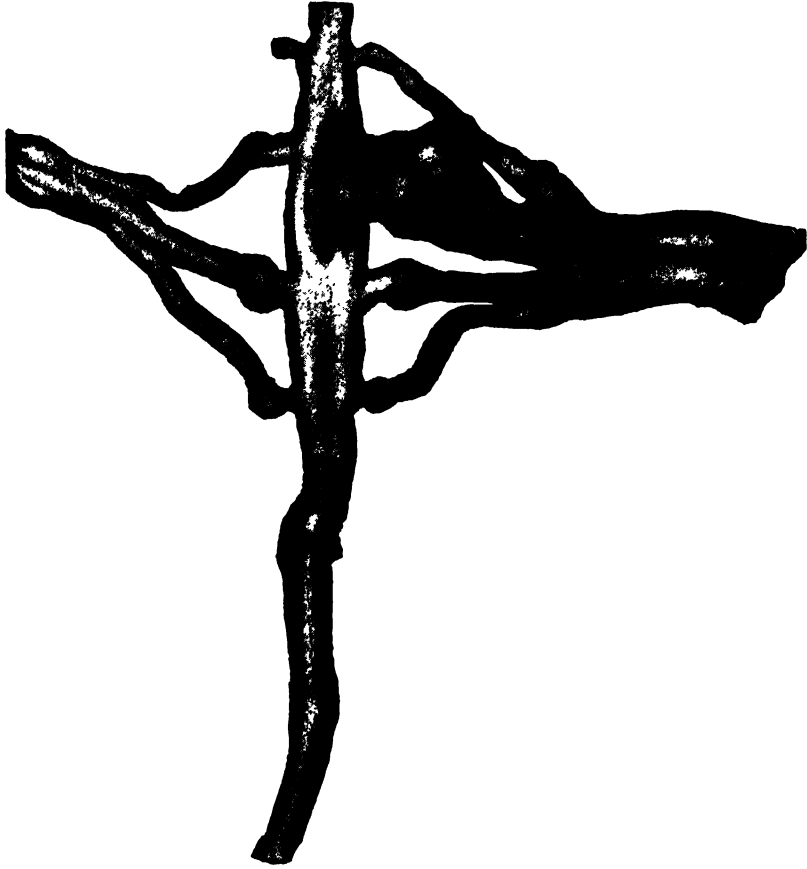


FIG. 18.4. Neural lymphomatosis. Dissection of spinal cord and brachial plexus. (Pappenheimer, Storrs Agr. Exper. Sta., Bul. 143.)

inflammatory lesions which were followed by the familiar lymphomatous changes. Glover (1940) made similar observations in chicks experimentally inoculated with neurolymphomatosis from pheasants. Asplin (1944) made the interesting observation that a "chick disease" of known viral etiology was amenable to sulfa drug therapy. In further studies he (Asplin, 1947a) confirmed the frequent association of the "chick disease" virus with field cases of lymphomatosis but failed to substantiate any etiologic connection with lymphomatosis (1947b).

On microscopic examination affected nerves present either follicular or diffuse infiltration with mononuclear cells (Fig. 18.5). The majority of the pathologic elements are indistinguishable from lymphocytes in the circulating blood and correspond to so-called small round cells; others have the character of plasma cells, large mononuclears, or histiocytes. The lesions may be



FIG. 18.5. Neural lymphomatosis. Section of sciatic nerve. (Pappenheimer, Storrs Agr. Exper. Sta., Bul. 143.)

associated with edema, myelin degeneration, and reactive increase of the Schwann sheath cells, but axonal degenerations are rare. Spinal and sympathetic ganglia undergo similar infiltrative changes. The disease also affects the central nervous system where it brings about either compact perivascular rings of small densely staining lymphoid cells or submiliary nodules composed of such cells and paler elements, probably of glial origin (Pappenheimer et al., 1926). Granulocytic reactions are not characteristic. The in-

tensity of the nervous system lesions seems to vary inversely with their development in the peripheral nerves. Ordinarily, involvement of the central nervous system is secondary to that of the peripheral nervous system, but one has to recognize the occurrence of a "central" variety of neural lymphomatosis not associated with peripheral nerve lesions. In a recent case of "torticollis," the auditory nerve presented the principal lesions.

Microscopic lesions frequently occur in grossly normal-appearing nerves. Microscopic examination thus constitutes the most sensitive method of diagnosis available at the present time. Due to the focal distribution of the lesions, the accuracy of the method is proportional to the number of segments of the nervous system examined. Standard histologic methods including frozen section techniques are applicable; in experienced hands, impression smears give quick diagnostic results, but are not reliable in mild cases.

**Hematology.** In a basic publication Pappenheimer *et al.* (1926) stated that, although the blood was not subjected to a systematic study, no indications of significant blood changes were observed. Absolute and relative lymphocytosis was noticed by Bayon (1931) in four acute cases. Johnson and Conner (1933) reported an absolute increase in leukocytes, a relative increase of monocytes and basophils, and the appearance of "budded" lymphocytes. Observing an early polymorphonuclear and late lymphocytic leukocytosis, Seagar (1933) made this the basis of the so-called cyto-diagnosis of fowl paralysis. Blount (1934), Dobson (1934), Hamilton (1934), and Blakemore (1934) pronounced this test unreliable and nonspecific. Gibbs (1934) confirmed the observations of Seagar, but considered the blood changes of little diagnostic assistance, on account of normal hematologic irregularities. In his pathologic characterization of fowl paralysis. Furth (1935) pointed out the usual lack of blood and bone marrow involvement. Jungherr (1934) made serial hematologic examinations of experimentally injected and control birds; the mild evidence of leukocytosis and lymphocytosis found in affected birds was regarded to be statistically insignificant. Incidentally, it was brought out that laboratory-kept young birds show an accentuation of the normal lymphocytic character of the avian blood.

**Differential diagnosis.** As has been stated, other morbid conditions summarized under the term symptomatic paralysis (Bayon, 1932) may be accompanied by paretic signs. In questionable cases the specific diagnosis of neurolymphomatosis rests upon the microscopic demonstration of the pathognomonic lesions in the peripheral nerves. Brooder chicks may show in the sciatic trunks, as in other organs, slight granulocytic mononuclear foci which are probably expressions of ectopic hematopoiesis and of no diagnostic significance. Neuromalacia or ariboflavinosis (Phillips and Engel, 1938) is characterized by bilateral diffuse grayish swelling of the sciatic trunks, which on microscopic examination exhibit severe myelin degeneration and occa-

sionally mild perivascular proliferation of the adventitial cells. The microscopic lesions of central neurolymphomatosis and avian encephalomyelitis (Jones, 1934) are similar in character except that the latter have a special predilection for the gray matter in the brain (Jungherr, 1935). The so-called "crooked-toes" condition described by Norris *et al.* (1940), apparently related to mechanically unsuitable floors, is not associated with nerve alterations. Intracranial gliomas may occasionally give rise to paralytic symptoms (Jungherr and Wolf, 1939).
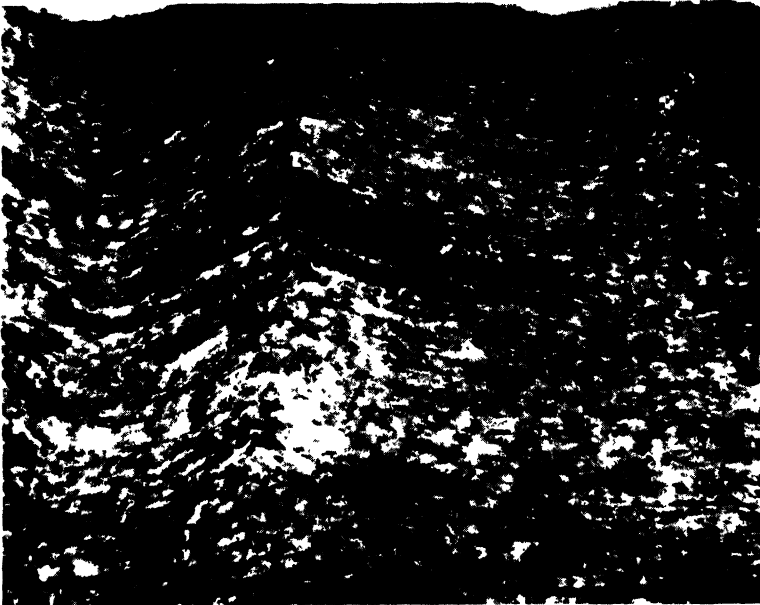


FIG. 18.6. Neural lymphomatosis in pheasants. Section of sciatic nerve.

### NEURAL LYMPHOMATOSIS IN OTHER SPECIES

A disease pathologically indistinguishable from the corresponding condition in the common fowl has been observed in pheasants by Jungherr (1939) (Fig. 18.6). An unusual feature was the finding of localized areas of muscle degeneration, especially in the flexors of the leg which led to macroscopic "tigering" of the affected muscle and microscopic Zenker's degeneration associated with regenerative phenomena (Fig. 18.7), similar to the muscle changes sometimes observed in young chickens by Potel (1938). Harriss (1939) and Johnson (1941) apparently transmitted fowl paralysis to pheasants. A similar affection of both neural and muscular tissues has been described in turkeys by Andrewes and Glover (1939), who regarded it as representing true fowl paralysis in another species of bird. Glover (1940) transmitted turkey paralysis to chicks and believed it to be caused by the same agent which is responsible for chicken paralysis.

## OCULAR LYMPHOMATOSIS

**Synonyms.** Blindness, gray-, glass-, pearly-, fish-eye, iritis (Pappenheimer *et al.,* 1926) ; epidemic blindness (Findlay and Wright, 1933) ; uveitis (Doyle, 1928) .

As already noted, Kaupp (1921) , Doyle (1926) , and Pappenheimer *et al.* (1926) have observed the frequent association of blindness with cases of neural lymphomatosis. Findlay and Wright (1933) and Jaensch and Lerche (1933) believed epidemic blindness to be an expression of fowl paralysis.
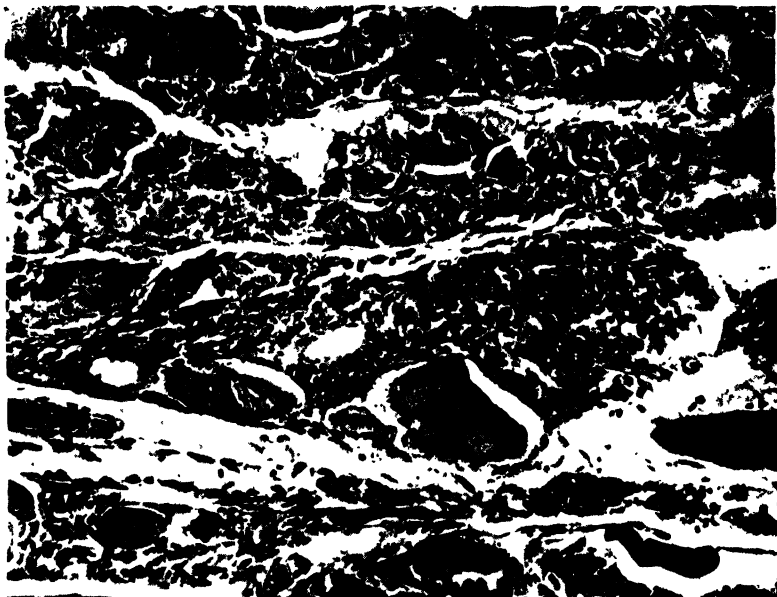


FIG. 18.7. Neural lymphomatosis in pheasants. Section of leg muscle showing Zenker's degeneration.

Similar epidemiologic observations were made by De Boer (1934a, b) in Holland, and Magnusson (1935) in Sweden. Although Upp and Tower (1936) , on the basis of crossbreeding studies with paralyzed and blind birds, regarded blindness as an independent disease, later studies by McClary and Upp (1939) showed a high incidence of iritis in the progeny of iritis-affected parents. Bayon (1936) gave a pathologic description of a primary irido-cyclitis in fowl said to be distinct from ocular lymphomatosis. Starting with affected iris material, Jungherr (1937) apparently developed transmissible strains of neuro-visceral lymphomatosis. Probably the most elaborate breeding experiments with birds of which either one or both parents had ocular lymphomatosis were carried out by Lee and Wilcke (1941) , who found that the offspring showed a significantly higher incidence of various forms of the avian leukosis complex than that of nonaffected control stock. Nelson and

Thorp (1943) found pearl-gray irises with irregular pupillary borders to be extensively infiltrated with lymphocytes. They believed the early stages of the disease, without pupillary changes, to be represented by depigmentation and vascular congestion of the iris. Using chicks from iritic dams as virus donors, Durant and McDougle (1945) transmitted neural lymphomatosis by direct blood transfusion to susceptible chicks.

On the other hand, Ball (1944) working with Single Comb White Leghorns found depigmentation of the iris, with round regular pupils, to be common and due to various factors such as lack of carotenoid pigments in the diet, and high egg production. A large proportion of such depigmented irises showed lymphocytic infiltration on microscopic study (Ball, 1945). Breeding from birds so affected failed to indicate a significant relationship between iris color of the dam and mortality from neoplastic or other diseases in her progeny (Ball and Cole, 1946).

**Occurrence.** Ocular lymphomatosis occurs primarily in flocks affected with the neural form but, on the whole, somewhat later during the life of the birds, primarily during early maturity. In a few cases it has been observed at the age of four weeks. Some outbreaks are not associated with other manifestations, and might lure the casual observer into a false sense of security regarding the avian leukosis complex status in the flock.

**Symptoms.** Since the ocular form constitutes the principal clinical evidence of latent affection with the avian leukosis complex, familiarity with the symptomatology is necessary for all who are concerned with the selection of breeding birds. Refinements in clinico-diagnostic methods, perhaps with the aid of an ophthalmoscope, should be helpful.

In the normal eye the iris has either a clear bay or orange color; the pupil is circular and has the power of ready accommodation to light intensity. The iris sometimes shows fine radial lines or clefts, which are probably in the nature of congenital defects.

Ocular lymphomatosis manifests itself by concentric annular or spotty depigmentation or by diffuse bluish-gray fading of the iris, in one or both eyes (Fig. 18.8). The pupil becomes irregular to the extent of showing angular indentations and gradual loss of light accommodation. In advanced stages the iris presents a diffuse grayish opacity with pin-point pupil (Fig. 18.9). The anterior chamber of the eye sometimes contains slightly turbid exudate which leads to increased convexity of the cornea as in glaucoma.

Considerable experience is needed in judging eyes as to normality or abnormality. From the practical standpoint it is well to err on the side of severity, in culling birds for significant eye lesions because "one abnormal eye is as bad as two."

**Pathology.** The specific pathologic features of the disease can be demonstrated only by histologic methods. Both eyes and brain should be examined.

The iris is most frequently affected and shows mononuclear infiltrates usually consisting of small round cells occasionally mixed with large polyblast-like cells (Fig. 18.10). Exclusive infiltration with heterophils is not characteristic, and raises the question of traumatic ophthalmitis. The anterior chamber of the eye may contain granular or amorphous material (Fig. 18.11); actual cellular exudate is rare. The bulbar lesions may extend into the eye muscles, especially the *Rectus lateralis* and *ciliaris*. In a comparatively small number of cases, lesions are observed in the cornea near Schlemm's canal, bulbar conjunctiva, pecten, retina, and optic nerve (Findlay and Wright, 1933; Gibbs and Johnson, 1936). The retina is sometimes separated from the choroid by lymphoid, follicle-like structures. Hepding (1939)



FIG. 18.8. Ocular lymphomatosis. The eye on the left shows annular, that on the right, spotty, depigmentation of the iris.

found accidental Toxoplasma infection in an affected retina. The optic nerve may show sparse infiltrates either close to the retina or more frequently in its intracranial course where it forms the prominent optic tract. Some cases of encephalitic affection of the optic nerve are not associated with bulbar lesions; although there may be impaired vision, the eyes appear clinically normal.

**Hematology.** In uncomplicated cases of ocular lymphomatosis the blood picture does not deviate from the normal.

### VISCERAL LYMPHOMATOSIS

**Synonyms.** Big liver disease, lymphatic leukosis (Ellermann, 1922), visceral lymphomata (Pappenheimer *et al.*, 1926), lymphocythaemia or myelolymphomatosis (Bayon, 1930), hemocytoblastic myelosis (Battaglia and Leinati, 1929), lymphadenoma (Mathews and Walkey, 1929), lymphocytoma (Feldman, 1932), lymphomyelosis (Kitt, 1931), lymphomatosis (Johnson, 1932), lymphomatosis (Furth, 1933), hepato-lymphomatosis (Furth, 1935), lymphosarcoma, lymphocytomatosis, certain endothelioma (Furth, 1933).

The restricted conception of Pappenheimer *et al.* (1926) that visceral lymphomata originating in the ovary may be a manifestation of fowl paralysis was extended by Johnson (1932) to include the majority of the visceral lymphomata, regardless of their anatomic origin, and led him to suggest the term "visceral lymphomatosis," in which sense it is used here. Mathews and Walkey (1929), Feldman (1932), Fenstermacher (1932, 1934,

1936), and others considered the lymphoid tumors of the fowl to be true neoplasms for which transmissibility had not been demonstrated and which were to be separated from the probably infectious entity of neurolymphomatosis. Furth (1935) demonstrated certain pathologic and etiologic differences between his transmissible strains of lymphomatosis, and thereby gave a somewhat different meaning to the term lymphomatosis. Transmissible strains developed from ordinary field cases by Jungherr (1937) and others showed considerable overlapping between the differential characters of



Fig. 18.9. Ocular lymphomatosis. Uveitis causing total blindness. (L. P. Doyle, Jour. A.V.M.A.)

neurolymphomatosis and lymphomatosis Furth. Papers on fowl paralysis by Oakley (1935), Dalling and Warrack (1936), Harriss, Johnston and Mitchell (1947), Gildow *et al.* (1940), and others seem to have used the term lymphomatosis in the present sense.

**Occurrence.** There hardly can be any doubt that under the conditions of modern intensive poultry production, visceral lymphomatosis represents the most common type of avian neoplasia. Epidemiologically its occurrence goes parallel with the neural and ocular forms. However, one gains the impression that the visceral form occurs somewhat later than the nerve or eye type,

especially during the laying period of pullets. The malady has been recognized as early as the fourth week of the brooding period. The extreme affections of the liver, so-called hepato-lymphomatosis (Furth, 1935) seem to be reserved for mature pullets or hens.

**Symptoms.** Unless associated with the neural or ocular form, the outward signs of the disease are indefinite. The comb becomes pale and shrivelled, occasionally darkened to the point of cyanosis in liver involvement. The symptoms may be accompanied by loss of appetite, loss of flesh, or diarrhea.
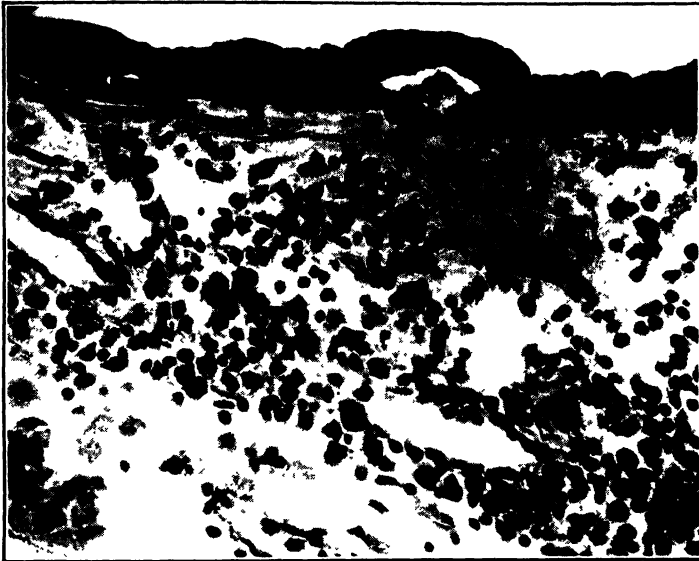


Fig. 18.10. Ocular lymphomatosis. Experimental case. Iris showing large- and small-cell lymphocytic infiltration. (Jungherr, Storrs Agr. Exper. Sta.)

Widespread attack of the mesentery can result in secondary ascites ("abdominal dropsy") and cause changes in contour and posture, spoken of as "penguin position." In systematic culling, sufficiently thin birds permit palpation of certain internal organs; thus, enlargement of the liver may be recognized by its projection beyond the metasternum and caudal margin of the ribs. The condition is often of long standing, but the observed clinical course may be quite short in that seemingly healthy birds succumb within a few days.

**Pathology.** Probably no other disease of birds presents a greater variety of gross-pathologic pictures than visceral lymphomatosis. The large abdominal glands, such as liver and kidney, are principally affected, but there is really no organ of the body, including the skin, which is not implicated at times.

In a consideration of the gross alterations, one might commence with the

spleen, because this organ is characteristically enlarged in most cases, up to three times the normal size. Splenic hyperplasia, however, may terminate in exhaustion and atrophy. The affected spleen is usually of grayish-brown color associated with milky thickenings in the capsule. Cross section exhibits minute grayish areas which correspond to hyperplastic lymphocytic aggre-



FIG. 18.11. Ocular lymphomatosis. Section through eye showing granular coagulum in anterior chamber, and cellular infiltration of iris. (Pappenheimer, Storrs Agr. Exper. Sta.)

gates. Instead of such diffuse changes, the spleen may present circumscribed projecting grayish tumors.

In the liver lymphomatosis likewise manifests itself in a "diffuse" and a "nodular" variety, according to Feldman (1932). Both may be present in the same specimen. In the former variety, which is the more common, the liver is enlarged to various degrees, grayish, and of granular surface (Fig. 18.12). The discoloration is often relieved by the red lines or dots of congested vessels which give the liver a characteristically marbleized appearance. In the

nodular variety there is less enlargement, but the parenchyma is studded with firm grayish tumors of various sizes, which unless confluent, are spherical except for surface flattening. On section the tumors are firm, smooth, lardaceous, and rarely show necrobiotic changes.

Implication of the gonads, especially in the female, has been stressed repeatedly (Pappenheimer *et al.*, 1926). The immature affected ovary may show nothing more than diffuse coarse-granular hyperplasia, while the producing organ exhibits alternating normal and tumorous egg follicles. Extensive affection is indicated by cauliflower-like tumors with multiple pedunculation. That such ovarian involvement may proceed at a rapid pace is indicated by birds which were observed to have a monthly trap nest record of twenty-four eggs, one month prior to death from ovarian lymphomatosis. Male gonads are sometimes subject to lymphomatous affection, the disease having been diagnosed as early as the fourth week of age. Marked differences in the size of the testicles should arouse suspicion of the disease, which is to be confirmed microscopically. Veritable tumor formation in the testicles is rare.

Among other internal organs the kidneys are often involved, primarily by diffuse grayish enlargement of some of the three major



FIG. 18.12. Visceral lymphomatosis, diffuse variety. The liver is markedly enlarged, grayish, and of granular surface. (Iowa State College.)

lobes, although nodular tumor formations occur at times. Due to the decrease of the renal secretory surface, affected kidneys may show secondary nephritic changes of the nonaffected lobes. Without concomitant lymphomatous alterations, these kidney lesions may be difficult to differentiate from uremic nephritis (visceral gout).

The heart shows gross changes more frequently than is realized; there occur either grayish striations (tigering) in the epicardium or prominent myocardial tumors which lead to deformities and local adhesive pericarditis; at times, the bicuspid valve presents small tumorous nodules. The lympho-

matous process is also apt to originate in or spread to adjacent organs such as the proventriculus, gizzard, and lungs (Fig. 18.13). Multiple tumors are often implanted in the mesentery and peritoneum, recalling the appearance of "pearl disease" in bovine tuberculosis. In such cases there is apt to be serous transudation into the abdominal cavity. Oviduct and mesosalpinx are comparatively refractory.



FIG. 18.13. Visceral lymphomatosis. Marked tumorous involvement of both lungs. (F. D. Patterson, Iowa State College.)

The glands of the head and the skin are subject to attack by the process. Unilateral swelling of the cheek, as in "roup," may be due to lymphomatosis of the naso-lachrymal glands. Affected integument exhibits small multiple tumors of the feather follicles, or large skin tumors prone to superficial ulceration. Skin involvement often escapes detection until the birds are dressed.

The condition of the bone marrow is ordinarily analogous to that described in neural lymphomatosis (Johnson, 1934), but frank leukemic cases of lymphomatosis may show minute grayish tumors (Furth, 1933).

Endothelioma apparently is a rare type of tumor in visceral lymphomatosis. It was first described as a transmissible condition in the fowl by Begg (1927), and again observed in transmission experiments of lymphomatosis by Furth (1933) and Jungherr (1937). The latter author has occasionally observed spontaneous cases of lymphomatous and endotheliomatous affections on the same ovary. Gross combinations of edematous grayish and hemorrhagic tumors should make one suspect an endotheliomatous process; the specific diagnosis rests upon microscopic study.

In contrast to the gross manifestations, the histopathologic picture of visceral lymphomatosis is comparatively uniform. Typical changes are represented by massive accumulations in various organs of proliferating lymphoid cells which are usually much more widely distributed than is suggested by gross alterations. The pathologic elements are represented by two prototypes, namely, so-called small round cells resembling lymphocytes, and large lymphoblast-like cells. Between these extremes intermediate types occur. The small more mature cell type is apt to occur in visceral complications of the neural form and in the extreme cases of liver enlargement (Fig. 18.14), while the large cell type prevails in rapidly growing lymphoid tumors.

Difficulties may arise in the interpretation of microscopic lymphomatous changes. As would be expected, the lesions have a predilection for organs containing normal lymphoid tissue. With the exception of cecal tonsils, bursa of Fabricius, submucosa of the intestine, and of the secondary bronchi, there are no organized lymph nodes in the fowl; but microscopic lymphoid follicles are normally present without anatomic regularity in the parenchymatous organs and the digestive tract. These follicles are subject to stimulation, aside from the etiologic agent of lymphomatosis, also by bacterial and viral factors (e.g., avian encephalomyelitis). Thus it is often difficult to state whether a given lymphoid follicle represents normal, reactive, or neoplastic tissue. A guide to interpretation should come from the consideration of all the pathologic evidence available in the case. In ordinary fixed tissues the large lymphoblast-like cells are difficult to differentiate from myeloblasts, hemocytoblasts, etc. Special fixing and staining methods designed to bring out the differential characters of hematopoietic elements are of value. Recourse may be taken to impression smears or "Klatsch" preparations stained according to Wright and/or Giemsa.

Regarding the micropathology of individual organs, the spleen in early cases exhibits lymphoid hyperplasia in and around the "adenoid sheaths," which are the probable functional equivalents of the Malpighian corpuscles in mammals. The hyperplastic areas become confluent and form broad

anastomosing masses which compress the red pulp. The process apparently takes its inception from the white pulp. True Malpighian corpuscles are absent in the bird, but organized lymphoid follicles appear irregularly in the normal spleen. They likewise undergo hyperplasia and merge imperceptibly with the affected adenoid sheaths.

The liver is often involved in the absence of gross lesions. Miliary lymphoid accumulations appear in the parenchyma or the portal islands. In the latter location, as elsewhere, there is frequently a secondary admixture of
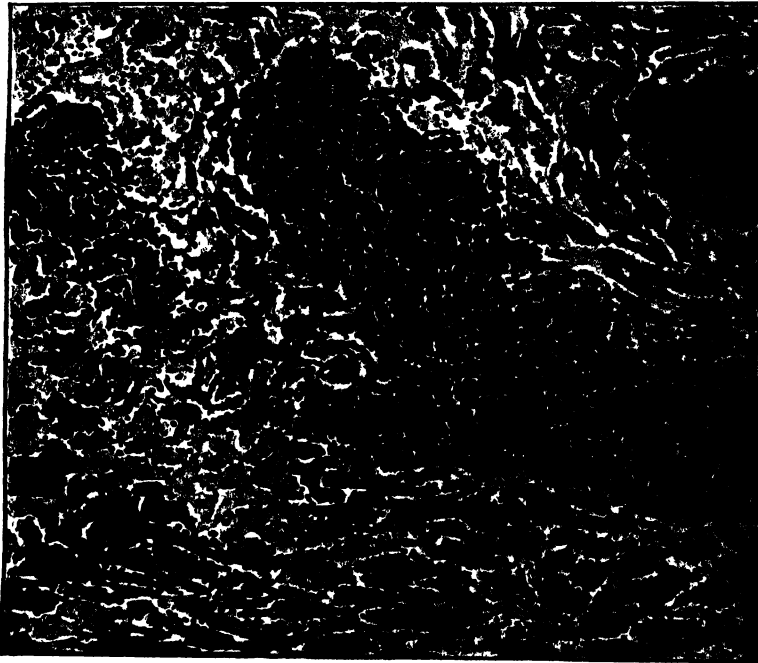


Fig. 18.14. Visceral lymphomatosis. Section of liver showing massive lymphoid infiltration. (Pappenheimer, Storrs Agr. Exper. Sta.)

metamyelocytes and heterophils, probably because such areas are still endowed with some of their embryonic hematopoietic potentialities. For severe cases massive compact expanses of lymphoid tissue are typical. They are in sharp relief from the surviving liver tissue by virtue of their basophilic staining affinity. The hepatic parenchyma itself shows only slight damage, a fact which may account for the surprising vitality of some birds in the presence of liver involvement.

The ovary shows either follicular or infiltrative lymphoid alterations; the latter often encroach upon or engulf ovarian follicles. In the kidney the process exhibits frequently an infiltrative interstitial character; since "interstitial nephritis" is rare in birds, such changes are often an expression of

lymphomatosis. The myocardium is particularly susceptible; in the beginning the lesions are infiltrative, but later form coherent tumorous masses. It is interesting to note that the cardiac muscle of healthy young birds often shows small ectopic foci of lympho- and granulopoiesis (Pappenheimer, Dunn, and Cone, 1929a). Although recognized now as part of the normal histo-anatomy of the chicken, it may be these foci which undergo neoplastic degeneration under the influence of the lymphomatosis agent.

Endothelioma is considered to be a possible extension of the microscopic
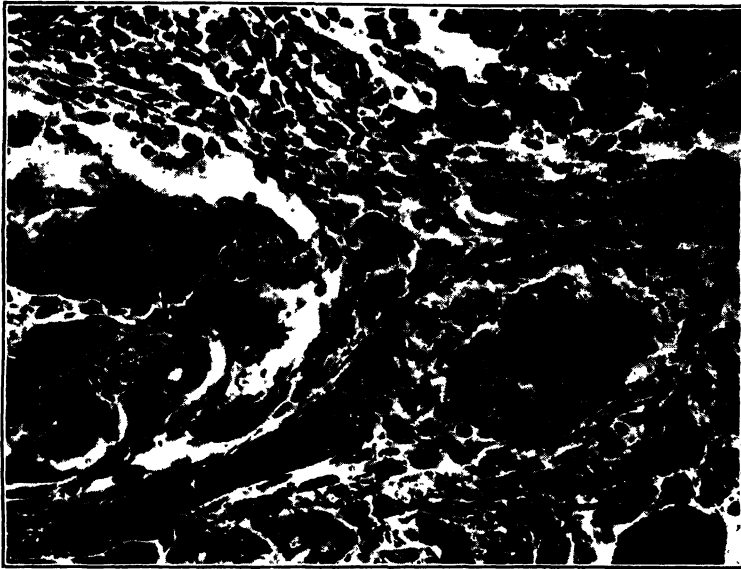


Fig. 18.15. Endothelioma in visceral lymphomatosis. Experimental case. Section of muscle affected with giant-cell endothelioma. (Jungherr, Storrs Agr. Exper. Sta.)

features of lymphomatosis. According to Furth (1934) neoplastic growth of endothelium may occur in any location of normal endothelium, including the liver, spleen, and bone marrow. Quite often these tumors are of purely microscopic size. The architecture is variable and may present solid, glandular, syncytial, and angiomatous types. The tumor is characteristically composed of basophilic cells with large chromatin-poor vesicular nuclei which exhibit thick nuclear membranes and prominent nucleoli. There occur frequently groups of multinucleated giant cells not unlike the Langhans type in tuberculosis (Fig. 18.15). The tendency toward necrobiotic changes is marked. The diagnosis of endothelioma is strengthened by demonstrating connection with normal endothelium. However, it is often difficult to decide whether the tumor is of endothelial, mesenchymal or mesothelial origin.

**Hematology.** Much of what has been stated under this heading in regard to neural lymphomatosis is applicable to the aleukemic cases of visceral

lymphomatosis. In other instances the blood picture may show subleukemic or leukemic lymphoid alterations. The term "subleukemic" is used in accordance with the suggestion of Furth (1933) to indicate a moderate increase of the leukocyte count, up to 100,000 per mm.³ of blood.

Specific blood changes are either qualitative or also quantitative in nature of which the former deserves special attention; they are often transitory or terminal and, on the whole, less constant than in erythro- and granulo-blastosis. Aside from the appearance of "budded" or pseudo-poded lymphocytes in increased number, the blood picture may be characterized by the presence of immature lymphocytes in the circulating blood. The degree of immaturity of the leukemic cell varies. According to Furth (1933), in sub-leukemic forms one observes primarily medium-sized basophilic lymphocytes showing vacuoles and azurophilic granules. The large lymphocyte predominates in frank leukemic cases. Apart from size these cells are distinguished by a large eccentric nucleus composed of spongy chromatin and a comparatively narrow mass of intensely basophilic cytoplasm. These cells were considered by Furth (1934) to be capable of producing erythroblasts, myelocytes, and lymphocytes, and thus correspond to the hemocytoblast in the terminology of Ferrata-Maximow. The lympholeukemic alterations may be accompanied by anemic changes, such as basophilic erythrocytes and erythroblasts; also erythrogonia suggestive of erythroblastosis occur occasionally.

**Differential diagnosis.** Of other diseases capable of forming tumor-like nodules in the visceral organs, tuberculosis and pullorum disease must be kept in mind. Avian tuberculosis also attacks primarily spleen and liver, but the nodules are usually yellowish, granular on the surface, and readily separable from the parenchyma of the affected organ. Pullorum nodules are found in the heart of adult birds, particularly males, but as a rule, are accompanied by inflammatory and congestive phenomena. In both diseases bacterioscopic and cultural tests can give further clues to the etiologic nature of the pathologic processes.

### OSTEOPETROTIC LYMPHOMATOSIS

**Synonyms.** Thick-leg disease, marble bone, akropachia ossea and hyperplastic ostitis (Reinhardt, 1930), hypertrophic osteitis (Reis and Nobrega, 1936), osteodystrophia fibrosa cystica (Gohs, 1934a, b), diffuse osteoperiostitis (Pugh, 1927), Paget's disease (Venkataraman, 1936), osteopetrosis gallinarum (Jungherr and Landauer, 1938).

Hypertrophic osteopathies of the fowl have received occasional mention in the literature (Reinhardt, 1930). Sporadic outbreaks of diffuse osteoperiostitis in male birds have been reported by Pugh (1927) in England, and Venkataraman (1936) in India. Brochet (1935) named several diseases as possibly leading to bone deformities, namely acromegaly, Paget's disease,

tuberculosis, gigantism, osteosarcoma and hypervitaminosis; he thought hypertrophic bone changes in birds to be secondary to respiratory infections or endocrine dysfunctions.

Experimental studies along this line are of special interest. Oberling and his associates (1933c, 1934) maintained birds in outdoor cages on a mineral-poor but otherwise adequate diet and observed lesions which showed a striking resemblance to human osteodystrophia fibrosa cystica associated with parathyroid hyperplasia. Gohs (1934a, b), on the other hand, produced a similar pathologic picture, save for parathyroid hyperplasia, together with leukemic conditions, by repeated injections of normal embryonic or X-rayed adult avian bone marrow. Furth observed occasionally cases of osteosclerosis in his transmission studies of the leukosis complex. Spontaneous cases pathologically resembling those of Gohs were described under the term osteopetrosis gallinarum (Jungherr and Landauer, 1938) and found to be transmissible. The agent could not be separated from that of lymphomatosis. While confirming the associated occurrence, Brandly, Nelson, and Cottral (1941) observed certain differences in epizootiology, and Burmester (1947a) in sedimentability, between osteopetrosis and lymphomatosis which, according to them, suggest etiologic differences. Periosteal proliferation in long bones of chicken embryos receiving intravenous injections of normal blood or serum was observed by Brandly (1941). The condition described by Thiersch (1944) under the name of osteopathia hyperostotica scleroticans multiplex infantilis following intravenous injection of embryos with human leukemia material seems to be of similar nonspecific character.

**Occurrence.** In comparison with other manifestations of the avian leukosis complex, the occurrence of osteopetrotic lymphomatosis is rare. Some field cases are sporadic in nature and often escape detection until the birds are dressed. This is borne out by observations of the United States poultry meat inspection service. Isolated epiornithic cases predominantly in males have been described. The geographic distribution appears to be widespread according to recent reports from Canada (Moynihan, 1943; Biely, 1943); Sweden (Magnusson, 1946); and South Africa (Coles and Bronkhorst, 1946). In a flock kept for experimental purposes the latter authors observed thirty-nine cases which were believed to show a familial incidence and to be due to a unifactorial recessive character, in support of the contention of Hutt (1932), who found two of nine birds from the same dam to be affected with such abnormal osteogenesis.

**Symptoms.** If the disease affects the metatarsi it can be detected on clinical inspection. Palpation of the long bones may reveal additional cases. In the beginning diseased leg bones, to the exclusion of the phalanges, show abnormal convexities or irregular thickenings in the diaphyseal or metaphyseal regions. The affected areas are hot to the touch, hard, and insensi-
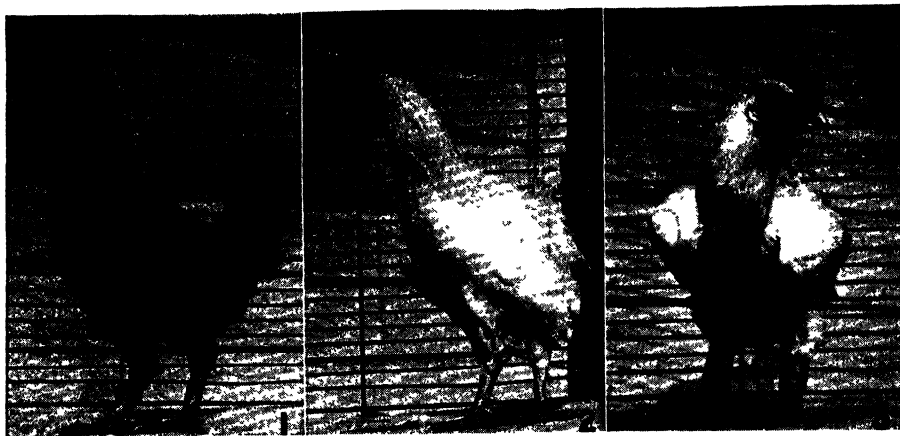
Fig. 18.16. Osteopetrotic lymphomatosis. The shanks of the birds are affected in various degrees. (Jungherr and Landauer, Storrs Agr. Exper. Sta.)

tive. Advanced cases exhibit the characteristic "boot-like" thickening of the shanks (Fig. 18.16) .

**Pathology.** The gross alterations of the skeleton may be observed in all the long bones of the extremities (Figs. 18.17 and 18.18), in the osseous components of the pelvis, shoulder girdle, and in rare instances also in the
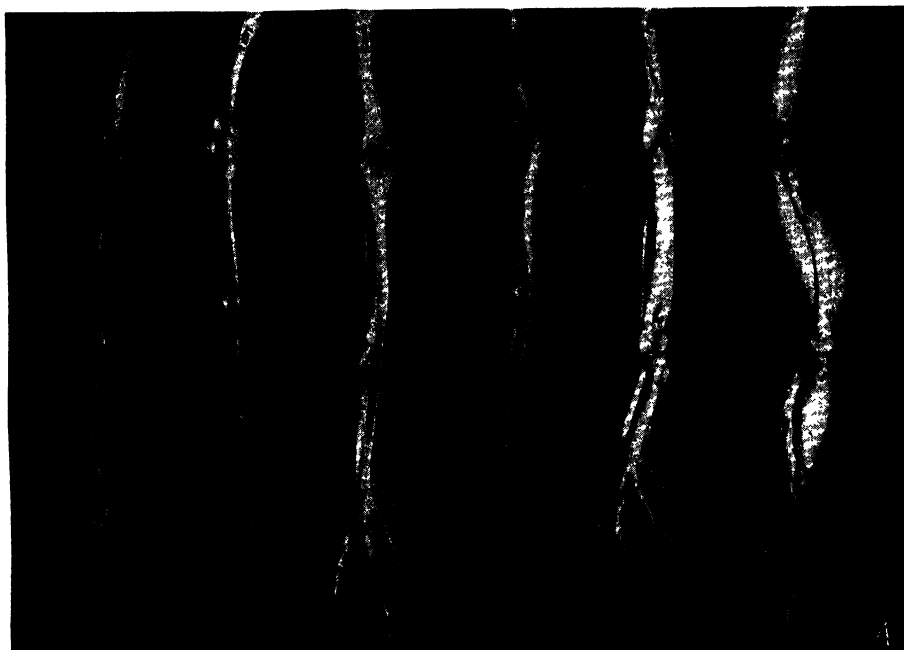


Fig. 18.17. Osteopetrotic lymphomatosis. Macerated leg bones affected in various degrees. First on left, normal. (Jungherr and Landauer, Storrs Agr. Exper. Sta.)

spine, while phalanges and skull bones seem refractory. X-ray pictures examined by Edeiken (1940), were considered to show definite bone pathology in the nature of osteosclerosis with thickening and increase in density of the cortex, encroachment and in some places obliteration of the medullary cavity (Fig. 18.19) ; the general features were regarded as similar to those of osteopetrosis in man.

The pathologic process affects primarily the diaphysis, and is ordinarily bilateral. The intensity of the bone lesions varies widely from exostosis-like
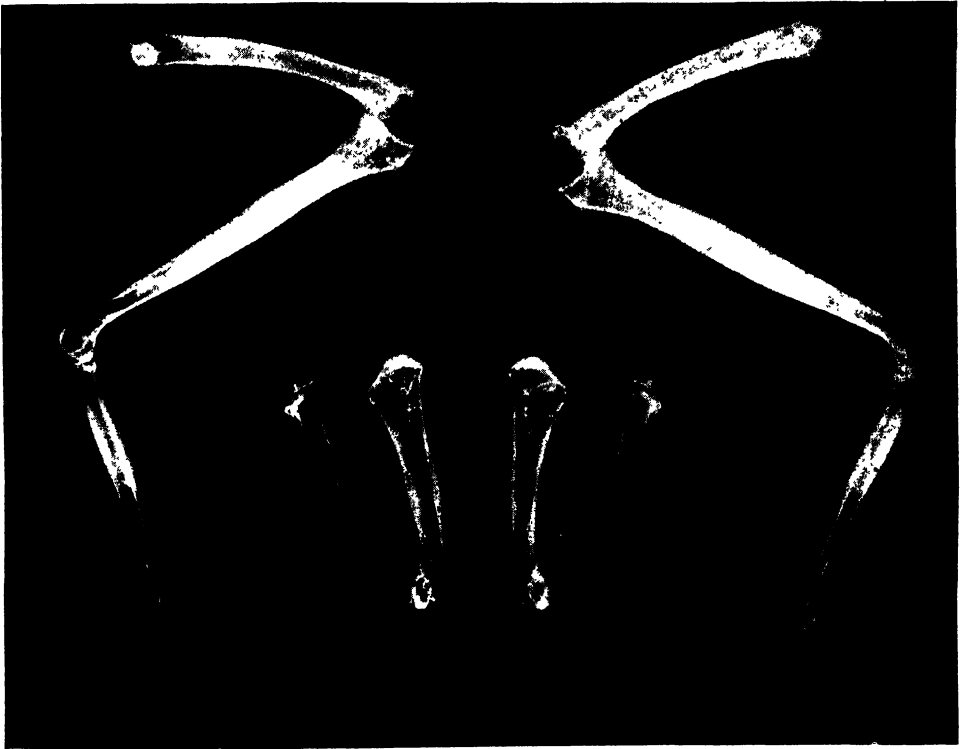


FIG. 18.18. Osteopetrotic lymphomatosis. X-ray picture of extremities from a 24-week-old light breed bird. (Vineland Poultry Laboratories, Vineland, N. J.)

cortical thickenings to massive asymmetrical involvement leading to almost complete obliteration of the marrow cavity. Even in the early stages the affected bones—though somewhat porous in appearance—give evidence of increased breaking strength. In cases of long standing they are extremely hard. Both cross (Fig. 18.20) and longitudinal sections of bones are helpful in the diagnosis.

The extent of the visceral pathology is variable. Rapidly developing cases usually exhibit the gross characteristic changes of visceral lymphomatosis,

while old arrested cases are more apt to show this association only on microscopic study of the tissues. The parathyroids appear normal.

The histopathologic picture of affected bones varies according to the maturation of the specific process. The initial phase is characterized by sequestration and granular degeneration of old trabeculae, marrow fibrosis, and increased osteoclasia. These changes, akin to those of osteodystrophia fibrosa, are accompanied by the development of a new large-celled vascular fibrous bone tissue which fails to show cartilage remnants or other evidence of endochondral ossification (Fig. 18.21). In the florid phase the new bone tissue gradually replaces both the original spongiosa and compacta, while osteoclastic activity regresses. In the arrested phase, the new bone lamellae appear condensed, hypercalcified and subdivided by numerous thick irregular cement lines (in formalin-fixed material) corresponding to the so-called "mosaic" structure of Paget's disease in man (osteitis deformans) (Fig. 18.22). All three phases may be observed in the same case or even the same bone section.



FIG. 18.19. Osteopetrotic lymphomatosis. X-ray picture of a White Rock chick about 9 weeks old, almost all long bones were involved. (Vineland Poultry Laboratories, Vineland, N. J.)

**Hematology.** The blood picture is ordinarily aleukemic. There is sometimes a relative or an absolute lymphocytosis. Evidence of secondary anemia is quite common and understandable, in view of the progressive reduction of the hematopoietic tissue. The remaining bone marrow is intensely hyperplastic, and on microscopic scrutiny shows basophil erythroblasts and erythrogonia, as in erythroblastosis. In spite of this hyperactivity of the marrow the immature stages of erythropoiesis have not been observed in the circulating blood.

**Differential diagnosis.** Among other avian osteopathies, rachitis and osteoporosis (rickets) can be differentiated from osteopetrosis by their epiphyseal formation of osteoid or porous bone tissue, accompanied in osteoporosis by parathyroid hyperplasia. Cases of osteopetrosis complicated by D-avitaminoses occur. In perosis (hock disease), there is twisting and flattening of the shanks, while the bone structure itself remains normal.

### ERYTHROBLASTOSIS

**Synonyms.** Severe anemia, "light," intravascular lymphoid leukosis (Ellermann, 1921), oligoerythrocythaemia or erythromyelosis (Bayon, 1929,
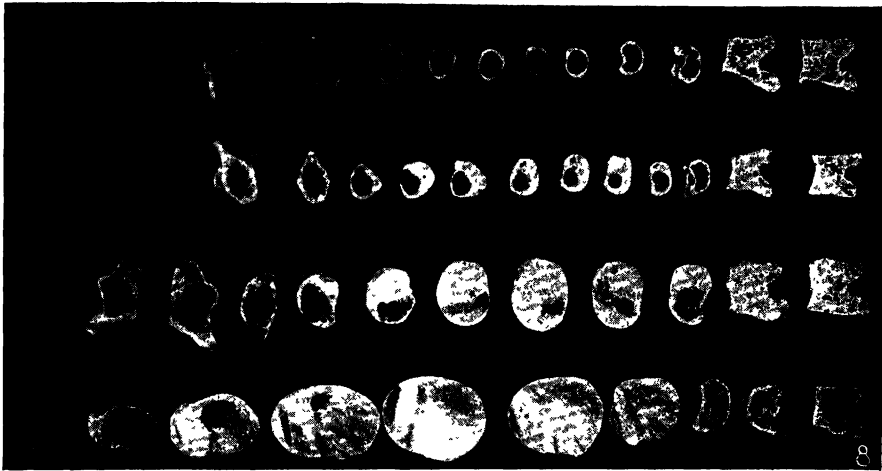


Fig. 18.20. Osteopetrotic lymphomatosis. Cross section of affected femora. Upper row normal. (Jungherr and Landauer, Storrs Agr. Exper. Sta.)

1930), erythroleukosis (Ellermann, 1923), (Furth, 1931b), leukomyelose (Kitt, 1931), the erythroblastic form of transmissible fowl leukosis (Olson, 1936), erythroblastosis (Engelbreth-Holm and Rothe Meyer, 1932).

Following the initial report by Ellermann and Bang in 1908 and subsequent studies (1921, 1923) on the experimental production of fowl leukemia by parenteral transmission of blood or blood-forming tissues, only Schmeisser (1915) reported on the successful transmission of a spontaneous case. As doubts had thus arisen in regard to the transmissibility of fowl leukosis, Jármai (1930) and Furth (1931a) independently developed new transmissible strains of fowl leukosis which proved to be of the erythroblastic and erythro-granuloblastic type, respectively. These results were soon confirmed by Engelbreth-Holm (1931) in Denmark, and Oberling, Guérin, and Boic (1933) in France, and others. From the pathologic standpoint most investigators recognized an erythroblastic and a granuloblastic (myeloid) type. Since both of them occurred in subpassages either alone or together,

irrespective of the disease in the donor, they were considered to be due to the same etiologic agent.

In transmission experiments on erythroleukosis, Ellermann (1923) observed severe cases of anemia and regarded them as hyperacute aleukemic expressions of the disease. Severe spontaneous anemia of fowl associated with lipochromatosis was considered to be different from Ellermann's erythroleukosis by Bedson and Knight (1924). By transmission of such cases, however, Stubbs and Furth (1932) were able to show their close relationship to erythroleukosis. Therefore, most investigators recognize that erythroblastosis may occur in a leukemic and an anemic subvariety (Olson, 1940).

**Occurrence.** In contrast to the amount of experimental work that has been conducted with it, erythroblastosis is of comparatively rare sporadic occurrence under field conditions. It is known to affect all standard breeds and occurs primarily after the age of six months (Olson, 1936). A notable exception is the report of Hamilton and Sawyer (1939), who observed the disease in five-week-old birds on an epiornithic scale.

**Symptoms.** While birds in the early stages of erythroblastosis appear normal, within a short time the disease manifests itself by paleness, or by a yellowish color of the unfeathered parts of the body, and by stupor and diarrhea; there is usually emaciation and loss of egg production. Uncontrollable bleeding from feather follicles has been observed at times. The anemic subvariety takes a chronic course over a period of several months.



Fig. 18.21. Osteopetrotic lymphomatosis. Section of metatarsus showing initial lesions. (Jungherr and Landauer, Storrs Agr. Exper. Sta.)

**Pathology.** The carcass may be emaciated, at times obese. Although the tissues appear pale, there are often petechial hemorrhages in various places such as the mucosa of the small intestine, under the liver capsule, or in the subcutaneous tissue. Terminally there is sometimes evidence of thrombosis, infarction, and rupture of internal organs. Edematous changes prevail in chronic cases. The most typical gross alteration is a diffuse enlargement of liver and kidneys and especially the spleen associated with a cherry-red

discoloration in fresh specimens. The parenchymatous organs are soft and friable. In uncomplicated cases there is no tendency toward the production of deforming tumors. Some of these changes may be absent, and even if they are well developed they are only suggestive and not diagnostic of the disease. Of special interest is the appearance in normally lipo-pneumatic long bones of the marrow which shows marked hemorrhagic hyperplasia and increased consistency described as "currant-jelly-like." The compacta of the long bones may undergo osteosclerotic changes (Bayon, 1930; Furth, 1931a).

In anemic cases of erythroblastosis the hyperplasia of the visceral organs and the bone marrow is absent; the spleen occasionally may be in a state of atrophy. The marrow spaces are often replaced to a large extent by a honeycombed spongy bone formation such as is seen in osteodystrophia fibrosa (Stubbs and Furth, 1932). Whether or not a given case of severe chronic anemia (asthenia in older textbooks) is causally related to erythroblastosis can be ascertained only by passage experiments.

On histologic examination, the lesions of erythroblastosis, aside from abnormal deposits of hemoglobin-derived pigments (Bayon, 1930), are characterized by dilatation of the blood sinusoids and latent and patent capillaries which are filled to capacity with primitive blood cells of the erythroblastic



Fig. 18.22. Osteopetrotic lymphomatosis. Section of metatarsus showing arrested lesions with "mosaic" structure. (Jungherr and Landauer, Storrs Agr. Exper. Sta.)

series. This essentially intravascular process is called leukostasis (Furth, 1931b) and is particularly developed in the liver, spleen, and bone marrow. Since mild hematologic changes suggestive of erythroblastosis may occur in other conditions, leukostasis is one of the prime features in the diagnosis of erythroblastosis.

In anemic erythroblastosis the visceral organs, especially the liver, often show extensive accumulations of small round cells and granulocytes which are probably reactive in character and difficult of interpretation. On careful search one is sometimes able to detect typical localized areas of leukostasis (Stubbs and Furth, 1932). According to Engelbreth-Holm (1932) the bone

marrow quite regularly shows microscopic accumulations of erythrogonia which, however, fail to invade the blood stream.

**Hematology.** Even on gross examination the blood is often noticed to be pale, watery, and slow to clot. As shown by Furth (1931a), centrifugalized specimens of fresh normal blood show a plasma:leukocyte:erythrocyte ratio of about 55:1:44; erythroblastic blood of 88:1:11; and granuloblastic blood of 16:69:15. Centrifugation of fresh blood in chilled tubes may thus serve as a presumptive diagnostic test, and in the case of erythroblastosis also gives evidence of severe anemia. In properly stained smears the blood picture is characterized by the appearance of many basophil erythroblasts and erythrogonia, which are considered to be hemoglobin-free precursors of erythrocytes. Although these cells are not strictly leukocytes, they would show up as leukocytes by the ordinary leukocyte count technique. The erythrogonia—which have been given the confusing name of lymphoidocytes by Ellermann—are highly characteristic; they vary in size, but are usually larger than the erythroblasts. In contrast to the "cart-wheel-like" nucleus of these cells, the nucleus of erythrogonia stains a peculiar violet red (in Wright-Giemsa preparations), and appears either homogeneous or diffusely granular. Binucleated and mitotic figures occur among them. The cytoplasm appears narrow, basophilic, and occasionally vacuolated (colored illustrations, Furth, 1931b). There is often an intense thrombocytopenia (Feldman and Olson, 1933). The number of polychrome erythrocytes is comparatively small. This is in contrast to the situation in secondary anemias—which may occur spontaneously or be induced by repeated bleeding or by certain chemicals—in which polychrome erythrocytes together with anisocytosis and poikilocytosis dominate the blood picture (Furth, 1931b).

**Differential diagnosis.** The danger lies not so much in mistaking erythroblastosis for another disease as in not suspecting it if it is present in an atypical or chronic form. Confirmation of the diagnosis must rest upon hematologic and histologic studies.

## GRANULOBLASTOSIS

**Synonyms.** Leukemic myeloid or myelotic leukosis (Ellermann, 1922), leukomyelose (Kitt, 1931), leucocythemia or leucomyelosis (Bayon, 1930), leukemic myeloblastosis (Nyfeldt, 1934), the granuloblastic form of transmissible fowl leukosis (Olson, 1936).

As has been emphasized by Furth (1934), the description of leukemic and aleukemic types of myeloid leukosis by Ellermann (1920) comprised two pathologically different conditions, namely myeloblastomatosis, which is identical with granuloblastosis, and myelocytomatosis. Aside from this, the historical development of available knowledge on granuloblastosis and erythroblastosis has been much the same, due to tendency of these diseases to

occur in a mixed form (Furth, 1931a). Nyfeldt (1934) claimed to have observed a pure strain producing only granuloblastosis.

**Occurrence and symptoms.** There are no essential differences between granuloblastosis and erythroblastosis. Bayon (1930) believes that the former occurs mostly in old hens.

**Pathology.** In distinction from erythroblastosis, the disease has a tendency to bring about grayish mottling of the enlarged parenchymatous organs; the bone marrow appears "pale or pink and diffluent" (Bayon, 1930). Otherwise, the anatomic changes are those described under erythroblastosis. Gross differential diagnosis between the two types is usually not possible (Olson, 1936). The histopathology of predominantly granuloblastic cases shows massive accumulation in the parenchymatous tissues of large myeloblastic and promyelocytic elements which are essentially extravascular, but overflow into the blood channels. There is therefore marked infiltration and substitution of the original tissues by the pathologic cells, in distinction from the fairly uniform intravascular leukostasis in erythroblastosis.

In the spleen the early changes seem to begin in the reticular stroma of the red pulp. The bone marrow shows intense granuloblastic activity in the extrasinusoidal areas, an observation which is particularly useful in pathogenetic studies. As may be surmised, the respective pathologic and hematologic features of erythro- and granuloblastosis show considerable overlapping in the mixed form.

**Hematology.** The marked rise in the leukocyte column of centrifugalized blood specimens has been mentioned (p. 456). The blood picture is characterized by the appearance of primitive cells of the granuloblastic series, especially myeloblasts (Fig. 18.23) and promyelocytes (colored illustrations, Oberling and Guérin, 1934) in large numbers, indicative of a leukemic or subleukemic state. The myeloblasts are large cells with slightly basophilic clear cytoplasm and a large vesicular acidophilic nucleus containing one to four nucleoli. Occasionally the leukemic elements are of the "Rieder" cell type (Olson, 1936). Promyelocytes and myelocytes have a similar nuclear structure, but can be definitely recognized by virtue of their specific granulation, which is primarily basophilic in the early forms, or grades into that of the three familiar types of adult granulocytes. Hemocytoblasts are indistinguishable from myeloblasts; if they occur together with definitely granulated elements, the former term seems to be more confusing than clarifying. In addition to the blood alterations mentioned, there may be evidence of secondary anemia. Late myelocytes and metamyelocytes with the granulation of the mature heterophil are rare.

## MYELOCYTOMATOSIS

**Synonyms.** Aleukemic myeloid leukosis (Ellermann, 1920), leukochloroma (Mathews, 1929), myelocytoma (Pentimalli, 1915; Feldman, 1932),

myelocytomatosis (Furth, 1933), aleukemic myeloblastosis (Nyfeldt, 1934), myeloma.

While it is difficult to state the exact nature of the disease described by Ellermann as aleukemic myelosis, Pentimalli (1915) first recognized the distinctive character of the tumor found in this disease, which Mathews (1929) described as an essentially aleukemic neoplasia. In passage experiments of lymphomatosis (Strain 2) Furth (1933) observed leukemic cases of myelocytomatosis and considered both to be pathologic expressions of the
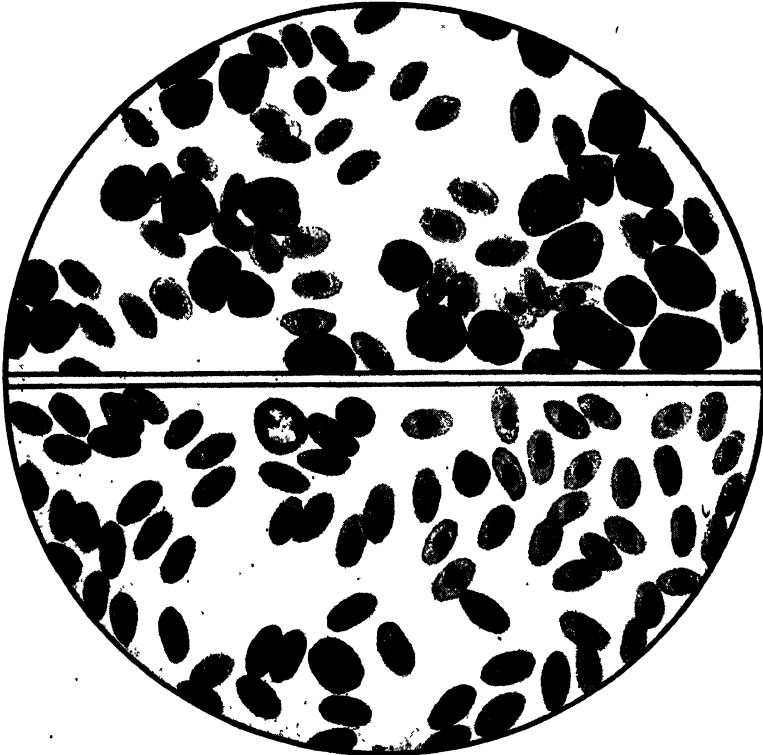


FIG. 18.23. Comparison of blood smears from (above) granuloblastosis and (below) neural lymphomatosis. (Pappenheimer, Storrs Agr. Exper. Sta.)

same agent. "Pure" transmissible strains with the possible exception of Nyfeldt's strain (1934), have apparently not been described in the literature. According to the available experimental evidence, myelocytomatosis is thus related to lymphomatosis, while everyday pathologic experience tends to place it closer to the ordinary tumors.

**Occurrence and symptoms.** Sporadic cases are apt to occur in young adult birds, some of which are seen in flocks that do not show any other pathologic evidence of the avian leukosis complex. The clinical appearance is noncontributory to the diagnosis.

**Pathology.** Myelocytomatosis characteristically forms tumors which figure among the few avian representatives which can be recognized on gross examination with some degree of certainty. The new growths are of yellowish-white color, resemble congealed cream in appearance and consistency, and are often of multiple anatomic origin. The masses may be found in the muscle tissue and visceral organs, and especially along the ribs or other parts of the skeleton bordering the body cavities, and may exert pressure on the spinal cord.

Histologically, the tumors consist of compact masses of myelocytes which are strikingly uniform in appearance and have the typical full acidophil granulation of either the mature eosinophil or the heterophil. The stroma is very scanty. In certain parenchymatous organs, such as the kidneys, the tumors show infiltrative growth. The bone marrow, as a rule, exhibits myelocytic hyperplasia. The type cell is ordinarily round, but may appear fusiform in areas subject to pressure.

**Hematology.** Uncomplicated cases are often aleukemic, although there may be a heterophil leukocytosis. If definite leukemic blood involvement occurs, the hematologic picture is dominated by the appearance of polychrome myelocytes and acidophilic metamyelocytes.

## ETIOLOGY

Discussion of the etiologic aspects of the avian leukosis complex presents difficulties which are brought about by the lack of definite information on the causal relationship of the various forms. Some points are of more or less general applicability.

Most observers are in agreement with the opinion of the early investigators (Marek, 1907; Pappenheimer *et al.*, 1926; Ellermann and Bang, 1908) that the condition is not caused by a cultivable organism of bacterial or fungal nature. Gray (1938) sometimes isolated Coccaceae from affected nerves.

On the evidence that field cases of fowl paralysis and allied conditions occur frequently in association with coccidiosis or helminthiasis, some students believe that parasitic conditions act as predisposing, precipitating, or even causal, factors. Bayon (1930), for instance, observed *Davainea proglottina* infection in erythroblastosis and severe anemia, and considered it to be a contributory factor, while Stubbs and Furth (1932), studying the same disease, failed to observe this association. The fact that typical cases of the various diseases of this group have been produced under controlled laboratory conditions (Warrack and Dalling, 1932), militates against a synergistic concept of the causation of the avian leukosis complex. On the other hand, the question of predisposing and aggravating factors must remain open and should best be attacked on an experimental rather than a statistical basis.

Some nutritional aspects were studied by Wilcke and his associates (1933), who found no significant differences in the incidence of fowl paralysis when the rations varied in mineral and vitamin levels. Blount (1932 interpreted this disease in terms of a B-hypervitaminosis or, more generally, of a gastronomic enteritis. On the basis of observing functional curative effects from the feeding of lettuce (Bayon, 1932), or a decrease in the incidence of the leukotic diseases following the feeding of additional wheat germ oil (Butler and Warren, 1938; together with Hammersland, 1938), some authors suggested the possibility of an underlying E-hypovitaminosis; the latter contention could not be supported by critical experimentation (Jungherr, 1940; Taylor and DeOme, 1939). Adamstone (1936), however, claimed to have produced a "lymphoblastoma"-like condition in chicks reared on a diet which had been treated with ferric chloride for the purpose of destroying vitamin E.

Nonspecific factors such as Salmonella toxins, vitamin A and K and iron deficiencies, poor ventilation, etc., have been claimed by Emmel (1939) to initiate blood changes which he termed hemocytoblastosis and considered to be the basic process in the development of the avian leukosis complex. McIntosh (1933) together with Selbie (1939) believed to have produced filtrable tumors in fowls by the injection of tar, but extended studies by Murphy and Sturm (1941a, b) on the transmissibility of chemically induced avian tumors so far have failed to confirm this observation.

The genetic aspects have been studied primarily on the basis of field observations. Thus Bayon (1932) voiced the widespread opinion that the incidence of fowl paralysis was higher in the progeny from certain strains of fowls. Asmundson and Biely (1932) and Biely, Palmer, and Asmundson (1932) found differences in the resistance to fowl paralysis, which they interpreted as due to a dominant inherited factor. Hutt (1939) and his associates (1941) observed reduction from 36 to 19 per cent in adult mortality due to neoplastic diseases after two generations of selecting disease-resistant lines. Cole (1941) developed White Leghorn resistant and susceptible lines for an artificially transmissible sarcoma (Jungherr, 1937), but found them to show no differences in spontaneous mortality which was principally due to lymphomatosis. This would indicate that genetic resistance to one type of neoplasia does not necessarily mean resistance to neoplastic disease in general. In ordinary observations it is difficult to differentiate between genetic susceptibility and actual transmission of the causative agent via the egg (as in pullorum disease). Thus it is seen that the genetic aspects are highly complex and intimately tied up with the unsettled questions of etiologic classification and gonadal transmission of the avian leukosis complex.

In recent times systematic studies have been undertaken to elucidate the

genetic factors governing resistance and susceptibility to lymphomatosis. By progeny-test selection over a period of eight years, Taylor et al. (1943) observed significant difference in the incidence of the disease between resistant and susceptible lines, but could not attribute them to sex-linked genes or to predominantly egg-borne transmission. The method of selection was believed to create a useful degree of relative resistance to lymphomatosis. On the basis of five years of selective breeding under conditions of rigid quarantine, Waters (1945b) demonstrated definite segregation of genes for resistance and susceptibility to lymphomatosis and emphasized the progressive nature of the process in contrast to previous workers who rarely achieved reduction of the disease below the initial percentage incidence. Differences between two sires became apparent when both were mated to the same dam (Waters, 1945c). In testing the resistance and susceptibility by artificial exposure, DeOme (1943) found intraperitoneal injections of lymphomatous nerve tissue to cause a more or less parallel increase of the incidence in both the high and low lines. By simulating natural exposure in the mouth, eyes, or nostrils, Heisdorf, Brewer, and Lamoreux (1947) obtained highly significant differences between resistant and susceptible lines, thereby pointing the way to further genetic approach to the control of lymphomatosis.

That sex hormone balance may influence the incidence of lymphomatosis and perhaps activate a latent agent, was suggested by Marine and Rosen (1941). Oakley (1935) already was impressed by the higher incidence of the visceral form in females than in males, while for the osteopetrotic form the reverse seemed to be true (Pugh, 1927; Brandly et al., 1941). The first-named authors observed a 50 per cent lymphomatosis incidence in a group of White Leghorn capons in contrast to none in the operative slips of the same lot; they then treated small lots of capons with sex hormones and found an incidence of 58 per cent in the estrogen-treated group and of 33 per cent in the androgen group, in contrast with 23 per cent in the controls.

These observations by Marine and Rosen were confirmed by Burmester (1945) who observed the incidence of natural lymphomatosis to be twice as high in females than in corresponding males during the first 300 days. In further studies along this line, Burmester and Nelson (1945) tested the influence of castration and of sex hormones and credited the male hormone with conferring increased resistance to lymphomatosis.

A transmissible agent[2] of fowl leukosis capable of passing through bacteria-retaining filters was first demonstrated by Ellermann and Bang (1908), and subsequently confirmed by many investigators (Jármai, 1930)

---

[2] The frequently made statement that the cause of the leukosis complex is unknown is essentially incorrect, as the etiologic agents can be identified by the methods which are peculiar to virology.

(Furth, 1931a, 1936b) (Thomsen and Engelbreth-Holm, 1932) (Oberling and Guérin, 1934). A similar etiologic agent was postulated for neurolymphomatosis by Van der Walle and Winkler-Junius (1924), Pappenheimer *et al.* (1926), Johnson (1932), and others, and for lymphomatosis by Furth (1933). These agents are similar to that of the Rous (Claude and Murphy, 1933; Foulds, 1934) sarcoma in being ultramicroscopic in nature and capable of producing a variety of neoplastic diseases in the fowl (Furth, 1932a). They have been termed microplasms, transmissible agents, enzymes (Jármai, 1935), or oncogenic viruses. These agents—as exemplified by the best-known erythroblastosis agent—resemble ordinary viruses in that they pass through all types of silicious filters and collodion membranes with a pore diameter of less than 400 m-microns (Johnson and Bell, 1936) (250 m-microns, Furth and Miller, 1932), are sedimentable according to Ledingham and Gye (1935), and have a particle size of about 72 m-microns according to Stern and Kirschbaum (1939); for a limited time they can resist careful desiccation, glycerination, and freezing by liquid air, and are thermolabile (Furth, 1932b). They cannot be cultivated except in the presence of viable cells (Furth and Breedis, 1937).

What set these agents apart as a group from ordinary viruses are certain properties outstanding among which is their neoplastic potentiality. On the whole, they are not only species-specific, but have selective affinities for certain tissues. However, both of these points are somewhat weakened by the findings of Jármai (1936) that pheasants, poults, and guinea fowl are .susceptible to the erythroblastosis agent of the chicken, and by those of Furth (1936a) that there occur "complex" leukosis-sarcoma strains. The agents seem to be present in high concentration in the blood cells and plasma (M.I.D. for erythroblastosis $10^{-5}$ to $10^{-7}$ cc. of blood) throughout the course of the disease (Furth, 1932a) but do not produce a solid immunity. Cellular inoculum usually produces more takes than cell-free material, regardless of the amount injected. The number of takes varies widely (25–70 per cent), as does also the incubation period which becomes especially prolonged if cell-free inoculum is used. Cultivation of the agents is possible only in the presence of such viable cells as are susceptible to the specific neoplastic potency of the agent (Furth and Breedis, 1937). The X-ray resistance is much higher than that which is tolerated by viable tumor cells or other "living" substances (Jármai, 1939). Although these agents can be concentrated, they cannot—by available methods—be isolated in a pure form, due chiefly to the fact that in the ultracentrifugal field they come down together with the macromolecular material present in normal tissues (Furth and Kabat, 1941). However, a difference from normal macromolecular material is demonstrable by neutralization tests (Kabat and Furth, 1941).

Whether or not the agents responsible for the leukosis complex and trans-

missible sarcoma constitute specific entities or variations of one and the same entity is the fundamental question, in arriving at an etiologic classification; this has been discussed in the historical part.

The principal claim as to the specificity of the agents is based upon the different pathologic responses engendered by them. That latent pathologic potencies may sometimes be uncovered by changes in the species and age of the host, however, was shown by Duran-Reynals, who produced a non-neoplastic "hemorrhagic disease" in chicks (1940) and ducklings (1941) that were injected with the agent of chicken Rous sarcoma. In older ducks the expected homologous disease was produced, but in the foreign host it lost its virulence for the donor species (chicken). Rous sarcoma virus that had been cultivated in vitro for ten years produced typical osteoid sarcoma on intravenous injection of growing chickens (Pikovski and Doljanski, 1946). While these results furnish food for thought, one has to recognize certain established differences among the transmissible agents of the leukosis complex. Under experimental conditions the agent of erythro-granuloblas-tosis usually affects birds of all ages, causes a high percentage of takes within an incubation period of about 30 days, and can withstand prolonged desiccation. According to Furth (1935) the causative factor of neuro-ocular lymphomatosis is transmissible only by viable cells. The filtrable agent of visceral lymphomatosis affects primarily young birds; the number of takes varies up to 56 per cent; it fails to produce tumors at the site of injection and, at times, produces osteopetrosis (Burmester, Prickett, and Belding, 1946a). However, exception to these norms are frequent.

An immunologic approach—such as is used in other virus diseases—to the question of interrelationship among the transmissible agents of the avian leukosis complex has been impeded by the frequent occurrence of normal resistant birds and by the usual lack of immunizing properties of the agents. Successful immunization against a transplantable lymphoid tumor did not increase resistance to naturally occurring neural or visceral lymphomatosis (Burmester, Prickett, and Belding, 1946b). Immunity reactions with various transplantable lymphoid tumors were constant toward the homologous, less so the heterologous, strains (Burmester and Belding, 1947). Chicken leukosis antiserums produced in ducks and turkeys exhibited a neutralizing effect toward chicken leukotic cells, often of a heterologous type (Lee, 1942). Preliminary complement fixation tests with turkey and guinea fowl immune serums were promising to aid in differentiation of the manifestations of the avian leukosis complex, according to Pollard, Hall, and Eichhorn (1943). Electrophoretic studies of the serum of normal and leukosis affected chickens disclosed in the latter a new "L component" fraction which may be of diagnostic significance (Sanders, Huddleson, and Schaible, 1944). Recently Kissling (1947) described a slide leuko-agglutination test with

formalinized lymphocytes from chickens or various mammals (except ox) for the diagnosis of avian lymphomatosis and believed the test to be primarily tissue and not tumor-specific.

## TRANSMISSION

· Because of the well-known difficulties encountered in the experimental transmission of the avian leukosis complex with blood and affected tissues, comparatively few studies have been reported on natural transmission, either through the egg, by contact, or by insects.

The earlier contentions of Doyle (1928) and McGaughey and Downie (1930) on the transmission of fowl paralysis through the egg did not find support in direct embryo inoculation experiments with this disease by McLennan (1935) or with erythroleukosis by Jármai (1933). Gibbs and Johnson (1935) claimed to have observed the characteristic pathologic cells of neurolymphomatosis in the follicular or seminal fluid of affected birds, while Storti and Mezzadra (1938) observed survival but not multiplication of the leukosis virus in 4-day-old chick embryos. Van den Berghe and d'Ursel (1939) apparently transmitted the leukosis agent to chick embryos, and Pollard and Hall (1941) to embryos of other avian species. Extensive breeding experiments by Lee and Wilcke (1941) have shown beyond a doubt that the incidence of the leukosis complex is much higher in the progeny from iritis-affected birds than from normal birds, especially if either the female or both parents were affected. Carrying on a similar breeding experiment, Durant and McDougle (1939) and the above authors were able to demonstrate the agent of fowl paralysis in the blood of recently hatched normal chicks. Field observations also support the thesis of dealing with an eggborne disease.

Until the reports of Patterson (1934), and Patterson et al. (1932), transmission of the avian leukosis complex by contact, e.g., by feces and contaminated litter, has been held improbable. Seagar's (1933) experiments, however, supported this contention, and so did the observations of Kennard and Chamberlin (1934), Hepding (1936), and others. Although Jármai and his associates (1932) found the natural body secretions devoid of the erythroblastosis agent, Jungherr (1937) and Fritzsche (1938) showed the occasional presence of the lymphomatosis agent in fresh and desiccated (one week) feces; Wagener (1939) and Blakemore (1939) considered the possibility of fowl paralysis transmission by contact. Gildow et al. (1940) believe that lymphomatosis is usually contracted by this means before the age of six to twelve weeks.

The question of transmission of lymphomatosis by contact and through the egg has been subjected anew to experimental inquiry. Barber (1942) reported a lower incidence of leukotic diseases in comparable groups of

birds reared away from, than on, known infected premises and was able to duplicate these results with resistant and susceptible stocks (Barber, 1943). In analyzing lymphomatosis mortality in sexually mature birds over a period of seven years, Hutt *et al.* (1944) observed a decreased incidence, independent from genetic background, in lots of birds that had been brooded for the first two weeks about 200 feet, in comparison with 40 feet for the controls, away from old birds. Such studies on the influence of the environment culminated in the development, by a system of selective breeding and extremely rigid sanitation, of a strain of birds essentially free from lymphomatosis by Waters and Prickett (1944). In follow-up work Waters (1945a) showed contact to be the principal mode of natural transmission of lymphomatosis and pointed out the importance of the egg as a carrier since simple importation of hatching eggs onto an entirely new isolated poultry plant resulted in the spontaneous appearance of lymphomatosis in chickens within 40 days after hatching. The unique contagious nature of lymphoid tumors in chickens, in contrast to most mammalian and avian tumors, was brought out in a recent discussion by Waters (1947).

Support for this thesis came from the work of Brewer and Brownstein (1947) in this country and Harriss *et al.* (1947) in England. Hutt and Cole (1947) questioned the importance of egg transmission of lymphomatosis, especially in comparison with genetic and environmental factors and referred to the work of Carr (1945) who failed to observe egg transmission in Rous sarcoma.

The presence of the avian leukoses agents in the blood stimulated research on blood-sucking parasites as intermediaries. While most of the experiments were negative (ref. Olson, 1940), Johnson (1937) showed the common red mite, *Dermanyssus gallinae*, and Brown and Cross (1941) the Texas "blue bug," *Argas persicus*, to be possible mechanical vectors of the lymphomatosis agent. Johnson (1937) also suggested that minor operations, as in fowl pox vaccination, may have the same effect.

## TREATMENT AND CONTROL

In general no practical therapeutic measures have been found for the avian leukosis complex. Neither the leukemic nor the anemic variety of erythroblastosis responds to iron or liver (Olson, 1936) treatment. The unconfirmed results with vitamin-E carriers have been mentioned (Butler and Warren, 1938). Recently parenteral injections of 10 per cent potassium iodide solution have been suggested for lymphomatosis by Gray (1940). It is to be remembered that temporary remissions of the clinical signs may occur spontaneously.

Vaccination attempts for the prevention of the avian leukosis complex have hardly gone beyond the exploratory stage. Fritzsche (1938) failed to

immunize birds against fowl paralysis with formol-treated tissue vaccine. Uhl (1938) secured some degree of immunity against erythrogranuloblastosis with aluminum hydroxide-adsorbed tissues. Johnson (1945) tried out several types of vaccines, usually consisting of formalinized tissues followed by unattenuated material intradermally, with inconclusive results.

Although a definite outline of a control program for the avian leukosis complex is not possible at the present time, the available facts suggest certain preventive measures.

1. In breeding stock, birds showing clinical evidence of the avian leukosis complex, especially true ocular lymphomatosis, should be consistently culled. Frequent laboratory checkups on the causes of mortality are advisable.

By systematic selection of the progeny for resistance to lymphomatosis, it is possible to obtain relatively resistant strains of birds (Taylor et al., 1943). In this it is preferable to choose breeders whose brothers and sisters have shown the lowest incidence of the avian leukosis complex (Gildow et al., 1940). Insofar as is known, the progeny of resistant stock remains fully susceptible (Blakemore, 1939). Although breeding from old birds is not in itself a guarantee of superior progeny, under practical conditions breeding from the survivors of natural selection and especially maintenance of a "closed flock" with a minimum of importations have given the most tangible results. Intentional enteral exposure to lymphomatosis, as tried by Heisdorf and his associates (1947), might be used in speeding up the segregation into resistant and susceptible breeders.

Although ordinary sanitary methods are generally conceded to be insufficient to prevent entirely the occurrence of avian lymphomatosis (Waters, 1945a; Harriss et al., 1947), hygiene as a factor in the control of the avian leukosis complex has been shown to be of major importance. Estimating the cost of fowl leukosis in the United States as $1,000,000 per week, Hutt (1944, 1945) proposes a simplified control program for the disease by brooding chicks for the first two weeks of life at considerable distance from adult fowl. Meticulous sanitary measures, based upon a conviction that lymphomatosis represents an infectious and contagious disease, have led to the establishment of the first flock of birds known to remain free from lymphomatosis for almost one year (Waters and Prickett, 1944).

A practical control program of the avian leukosis complex in the breeding flock must recognize the interaction of heredity and of infectious agents which may be both egg-borne and present in the environment. Such recognition postulates a selective breeding program for livability which will also significantly decrease the incidence of the avian leukosis complex (Bryant and Johnson, 1944), and a type of management that minimizes exposure to all types of infectious agents, particularly during the brooding and growing periods.

2. In non-self-sustaining flocks, chicks should be purchased from breeders who have adopted some or all of the above measures. Since, however, no tests are available for the detection of latent carriers of the leukosis complex in the breeding stock, the seller should not be held responsible for losses resulting from it.

Exacting sanitary and quarantine measures are advisable during the entire brooder stage and early maturity, to prevent transmission by contact and the possible precipitating effect of secondary parasitic factors.

3. Ectoparasites should be kept in check at all times. Minor operative procedures, such as vaccination, caponizing, etc., should be carried out with certain aseptic precautions, especially if the same surgical instruments are to be used on a series of birds.

## REFERENCES

Adamstone, F. B.: 1936. A lymphoblastoma occurring in young chicks reared on a diet treated with ferric chloride to destroy vitamin E. Am. Jour. Canc. 28:510.

Andersen, C. W., and Bang, O.: 1928. La leucémie ou leucose transmissible des poules. Festskrift til Prof. Bernhard Bang. Kopenhavn. Kandrup and Wunsch., p. 353.

Andrewes, C. H., and Glover, R. E.: 1939. A case of neurolymphomatosis in a turkey. Vet. Record 51:931.

Asmundson, V. S., and Biely, J.: 1932. Inheritance of resistance to fowl paralysis (neurolymphomatosis gallinarum). I. Differences in susceptibility. Canad. Jour. Res. 6:171.

Asplin, F. D.: 1944. Treatment of a virus disease of chickens with sulfonamides. Nature, London, 153:253.

———: 1947a. Observations on the aetiology of lymphomatosis. II. The association of "chick disease" virus with field cases of lymphomatosis. Jour. Comp. Path. and Therap. 57:126.

———: 1947b. Observations on the aetiology of lymphomatosis. III. The development of lymphomatosis in chickens free of the "chick disease" virus. Jour. Comp. Path. and Therap. 57:134.

Ball, R. F.: 1944. The effect of the ration upon iris color of Single Comb White Leghorns. Poultry Sci. 23:377.

———: 1945. A study of iris-depigmentation in Single Comb White Leghorns. Doctoral Thesis, Cornell Univ.

——— and Cole, R. K.: 1946. A study of the relationship between the iris color of the dam and the mortality of her progeny. Poultry Sci. 25:33.

Barber, C. W.: 1942. The effect of the rearing environment upon the incidence of avian leucosis complex. Cornell Vet. 32:191.

———: 1943. The effect of environment on the incidence of avian-leukosis complex lesions among resistant and nonresistant chickens. Cornell Vet. 33:78.

Battaglia, F., and Leinati, L.: 1929. Malattie sistemiche transmissibili degli organi emopoietici del pollo con ricerche sugli elementi morfologici del sangue normale e loro genesi. Boll. d. Inst. Sieroterap. Milanese. 8:9–31; 73–94; 183–98.

Bayon, H. P.: 1929. The pathology of transmissible anaemia (erythromyelosis) in the fowl. Parasitology 21:339.

———: 1930. The comparative pathology of anaemia and leucocythemia in fowls. Jour. Comp. Path. and Therap. 43:188.

———: 1931. Acute *neuro-lymphomatosis gallinarum* in a strain of Rhode Island Red fowls. Vet. Record 11:907.

———: 1932. The pathogenesis of *neurolymphomatosis gallinarum* and similar forms of "fowl paralysis." Vet. Record 12:457.

———: 1936. Primary irido-cyclitis in fowls: a condition distinct from the eye lesions occurring in neuro-lymphomatosis. Jour. Comp. Path. and Therap. 49:310.

Bedson, S. P., and Knight, E.: 1924. An anaemia in hens associated with an increase in the yellow pigment normally present in certain tissues of these birds. Jour. Path. and Bact. 27:239.

Begg, A. M.: 1927. A filterable endothelioma of the fowl. The Lancet 212:912.

Biely, J.: 1943. The avian leukosis complex. A note on avian osteopetrosis. Canad. Jour. Comp. Med. 7:276.

——— and Palmer, V. E.: 1932. The etiology of fowl paralysis (a review of the literature). Vet. Record 12:1302.

Biely, J., Palmer, E., and Asmundson, V. S.: 1932. Inheritance of resistance to fowl paralysis (*neurolymphomatosis gallinarum*). II. On a significant difference in the incidence of fowl paralysis in two groups of chicks. Canad. Jour. Res. 6:374.

Blakemore, F.: 1934. The leucocytes of fowl blood with special reference to fowl paralysis. Vet. Record 14:417.

———: 1939. The nature of fowl paralysis (neurolymphomatosis). Jour. Comp. Path. and Therap. 52:144.

——— and Dalling, T.: 1939. Some recent observations on fowl paralysis (neurolymphomatosis). Seventh World's Poultry Cong.. p. 282.

Blount, W. P.: 1932. Studies of fowl paralysis. III. Gastronomic enteritis. Vet. Jour. 88:236.

———: 1934. Fowl paralysis. Vet. Record 14:469.

———: 1939. Hemocytoblastosis. Vet. Jour. 95:91.

Brandly, C. A.: 1941. Progress report on several phases of pathology research. Rep. Second Collab. Conf. U. S. Reg. Poultry Res. Lab., East Lansing, Mich., 23:38.

———, Nelson, N. M., and Cottral, G. E.: 1941. Serial passage of strain 3, lymphomatosis-osteopetrosis in chickens. Jour. Am. Vet. Med. Assn. 99:219.

Brewer, N. R., and Brownstein, B.: 1946. The transmission of lymphomatosis in the fowl. Am. Jour. Vet. Res. 7:123.

Brochet, L.: 1935. Ostéite hypertrophiante chez la poule. Bul. Acad. vét. Fr. 88:191–96 and 477.

Brown, J. C., and Cross, J. C.: 1941. A probable agent for the transmission of fowl paralysis. Science 93:528.

Bryant, R. L., and Johnson, E. P.: 1944. Incidence of mortality in two strains of Single Comb White Leghorn chickens. Poultry Sci. 23:521.

Burmester, B. R.: 1945. The incidence of lymphomatosis among male and female chickens. Poultry Sci. 24:469.

———: 1947a. Centrifugation of a filtrable agent inducing osteopetrosis and lymphoid tumors in the domestic fowl. Poultry Sci. 26:215.

———: 1947b. Studies on the transmission of avian visceral lymphomatosis. II. Propagation of lymphomatosis with cellular and cell-free preparations. Cancer Res. (In Press.)

———: 1947c. Studies on the transmission of avian visceral lymphomatosis. II. Propagation of lymphomatosis with cellular and cell-free preparations. (Abst.) Poultry Sci. 26:534.

———: 1947d. The cytotoxic effect of avian lymphoid tumor antiserum. Cancer Res. (In Press.)

——— and Belding, T. C.: 1947. Immunity and cross immunity reactions obtained with several avian lymphoid tumor strains. Am. Jour. Vet. Res. 8:128.

———, Brandly, C. A., and Prickett, C. O.: 1944. Viability of a transmissible fowl tumor (Olson) upon storage at low temperatures. Proc. Soc. Exper. Biol. and Med. 55:203.

——— and Cottral, G. E.: 1947. The propagation of filtrable agents producing lymphoid tumors and osteopetrosis by serial passage in chickens. Cancer Res. (In Press.)

——— and Denington, E. M.: 1947. Studies on the transmission of avian visceral lymphomatosis. I. Variation in transmissibility of naturally occurring cases. Cancer Res. (In Press.)

——— and Nelson, N. M.: 1945. The effect of castration and sex hormones upon the incidence of lymphomatosis in chickens. Poultry Sci. 24:509.

——— and Prickett, C. O.: 1944. Immunity reactions obtained with a transmissible fowl tumor (Olson). Cancer Res. 4:364.

——— and Prickett, C. O.: 1945. The development of highly malignant tumor strains from naturally occurring avian lymphomatosis. Cancer Res. 5:652.

———, Prickett, C. O., and Belding, T. C.: 1946a. A filtrable agent producing lymphoid tumors and osteopetrosis in chickens. Cancer Res. 6:189.

———, Prickett, C. O., and Belding, T. C.: 1946b. The occurrence of neural and visceral lymphomatosis in chickens proven immune to transplants of lymphoid tumor strains. Poultry Sci. 25:616.

Butler, W. J., and Warren, D. M.: 1938. Fowl leukemia and vitamin E. Jour. Am. Vet. Med. Assn. 92:204.

———, Warren, D. M., and Hammersland, H. L.: 1938. Nutrition as a factor in the incidence of fowl leukosis. Jour. Am. Vet. Med. Assn. 93:307.

Carr, J. G.: 1945. Lack of transmission of avian tumour virus from carrier hens to their offspring via the egg. Proc. Roy. Soc. Edinb., Sec. B, 62:54.

Claude, A., and Murphy, J. B.: 1933. Transmissible tumors of the fowl. Physiol. Rev. 13:246.

Cole, R. K.: 1941. Genetic resistance to a transmissible sarcoma in the fowl. Cancer Res. 1:714.

Coles, J. D. W. A., and Bronkhorst, J. J.: 1946. The familial incidence of spontaneous osteopetrosis gallinarum. Onderstepoort Jour. Vet. Sci. and An. Ind. 21:79.

Dalling, T., and Warrack, G. H.: 1936. Observations on fowl paralysis (lymphomatosis). Vet. Jour. 92:310.

Davis, O. S., and Doyle, L. P.: 1947. Studies in avian leucosis. I. The transmissibility of visceral lymphomatosis. II. The use of biopsy technique in the study of visceral lymphomatosis. Am. Jour. Vet. Res. 8:103.

———, Doyle, L. P., Walkey, F. L., and Cenker, L. K.: 1947. Studies in avian leukosis. III. The incidence of avian leukosis in various breeds of chickens. Poultry Sci. 26:499.

De Boer, E.: 1934a. *Neurolymphomatosis gallinarum* I. Tijdschr. Diergeneesk. 61:449.

————: 1934b. *Neurolymphomatosis gallinarum* II. Tijdschr. Diergeneesk. 61:520.

DeOme, K. B.: 1943. Intraperitoneal injection of lymphomatosis nerve tissue into resistant and susceptible chickens. Poultry Sci. 22:381.

Doan, C. A., Cunningham, R. S., and Sabin, F. R.: 1925. Experimental studies on the origin and maturation of avian and mammalian red blood cells. Carnegie Inst. of Wash., Publ. 83:165.

Dobson, N.: 1934. Some poultry diseases. Vet. Record 14:332.

Doyle, L. P.: 1926. Neuritis in chickens. Jour. Am. Vet. Med. Assn. 68:622.

————: 1928. Neuritis or paralysis in chickens. Jour. Am. Vet. Med. Assn. 72:585.

Duran-Reynals, F.: 1940. A hemorrhagic disease occurring in chicks inoculated with the Rous and Fuginami viruses. Yale Jour. Biol. and Med. 13:77.

————: 1941. Age susceptibility of ducks to the virus of the Rous sarcoma and variation of the virus in the duck. Science 93:501.

Durant, A. J., and McDougle, H. C.: 1939. Studies on the origin and transmission of fowl paralysis (neurolymphomatosis) by blood inoculation. Mo. Agr. Exper. Sta., Res. Bul. 304.

———— and McDougle, H. C.: 1945. Further investigations of the transmission of fowl paralysis (neurolymphomatosis) by direct transfusion. Mo. Agr. Exper. Sta., Bul. 393:2.

Edeiken, L.: 1940. Private communication to Dr. A. D. Goldhaft, Vineland Poultry Laboratories, Vineland, N. J.

Ellermann, V.: 1920. Histogenese der übertragbaren Hühnerleukose. I. Die myeloische Leukose. Folia Haematol. 26:135.

————: 1921. Histogenese der übertragbaren Hühnerleukose. II. Die intravaskuläre lymphoide Leukose. Folia Haematol. 26:165.

————: 1922. The leucosis of fowls and leucemia problems. London, Gyldendal. P. 105.

————: 1923. Histogenese der übertragbaren Hühnerleukose. IV. Zusammenfassende Betrachtungen. Folia Haematol. 29:203.

———— and Bang, O.: 1908. Experimentelle Leukämie bei Hühnern. Zentralbl. f. Bakt. Abt. I. Orig. 46:595.

Emmel, M. W.: 1939. Hemocytoblastosis in the chicken. Proc. Seventh World's Poultry Cong.:298.

Engelbreth-Holm, J.: 1931. Bericht über einen neuen Stamm Hühnerleukose. Zeitschr. f. Immunitätsforsch. u. Exper. Therap. 73:126.

————: 1932. Untersuchungen über die sogenannte Erythroleukose bei Huhnern. Zeitschr. f. Immunitätsforsch. u. Exper. Therap. 75:425.

————: 1935. On the connection between erythroblastosis (haemocytoblastosis), myelosis, and sarcoma in chicken. Acta Path. et. Microbiol. Scand. 12:352.

————: 1942. Spontaneous and Experimental Leukaemia in Animals. Oliver and Boyd, London.

———— and Rothe Meyer, A.: 1932. II. Über den Zusammenhang zwischen den verschiedenen Hühnerleukoseformen (Anämie-Erythroblastose-Myelose). Acta Path. et Microbiol. Scand. 9:312.

Feldman, W. H.: 1932. Neoplasms of Domesticated Animals. W. B. Saunders Co., Philadelphia. Pp. 221–46.

———— and Olson, Jr., C.: 1933. The pathology of spontaneous leukosis of chickens. Jour. Am. Vet. Med. Assn. 82:875.

Fenstermacher, R.: 1932. Studies of leukemia of fowls. Jour. Am. Vet. Med. Assn. 80:791.

————: 1934. Familial incidence of lymphocytoma in three generations of the domestic fowl. Jour. Am. Vet. Med. Assn. 84:863.

————: 1936. Lymphocytoma and fowl paralysis. Jour. Am. Vet. Med. Assn. 88:600.

Findlay, G. M., and Wright, J.: 1933. Ocular lesions in epidemic blindness of fowls. Jour. Comp. Path. and Therap. 46:139.

Foulds, L.: 1934. The filterable tumours of fowls: A critical review. Sci. Rep. Invest. Imp. Canc. Res. Fund 11 (Suppl.). Pp. 1–41.

Fritzsche, K.: 1938. Versuche zur Erforschung und Bekämpfung der Marekschen Hühnerlähme. I. Versuche über die Ausscheidung des Hühnerlähmevirus mit dem Kot und über die natürliche Infektionsweise. Zeitschr. Infekt-Krankh. der Haustiere 52:51.

————: 1938. II. Versuch über die Brauchbarkeit des Präparates Therapeksi zur Bekämpfung der Hühnerlähme. Zeitschr. Infekt-Krankh. der Haustiere 52:124.

Furth, J.: 1931a. Observations with a new transmissible strain of the leucosis (leucemia) of fowls. Jour. Exper. Med. 53:243.

————: 1931b. Erythroleukosis and the anemias of the fowl. Arch. Path. 12:1.

————: 1932a. Studies on the nature of the agent transmitting leucosis of fowls. I. Its concentration in blood cells and plasma and relation to the incubation period. Jour. Exper. Med. 55:465.

————: 1932b. Studies on the nature of the agent transmitting leucosis of fowls. III. Resistance to desiccation, to glycerin, to freezing, and thawing; survival at ice box and incubator temperatures. Jour. Exper. Med. 55:495.

————: 1933. Lymphomatosis, myelomatosis, and endothelioma of chickens caused by a filtrable agent. I. Transmission experiments. Jour. Exper. Med. 58:253.

Furth, J.: 1934. Lymphomatosis, myelomatosis and endothelioma of chickens caused by a filtrable agent. II. Morphological characteristics of the endotheliomata caused by this agent. Jour. Exper. Med. 59:501.

———: 1936a. The relation of leukosis to sarcoma of chickens. II. Mixed osteochondrosarcoma and lymphomatosis (Strain 12). Jour. Exper. Med. 63:127.

———: 1936b. The relation of leukosis to sarcoma of chickens. III. Sarcomata of strains 11 and 15 and their relation to leukosis. Jour. Exper. Med. 63:145.

———: 1946. Recent experimental studies on leukemia. Physiol. Rev. 26:47.

——— and Breedis, C.: 1931. On the resistance and filterability of the agent transmitting leucosis. Proc. Soc. Exper. Biol. and Med. 28:985.

——— with assist. of Breedis, C.: 1935. Lymphomatosis in relation to fowl paralysis. Arch. Path. 20:379.

——— and Breedis, C.: 1937. Attempts at cultivation of viruses producing leukosis in fowls. Arch. Path. 24:281.

——— and Kabat, E. A.: 1941. Immunological specificity of material sedimentable at high speed present in normal and tumor tissues. Jour. Exper. Med. 74:247.

——— and Miller, H. K.: 1932. Studies on the nature of the agent transmitting leucosis of fowls. II. Filtration of leucemic plasma. Jour. Exper. Med. 55:479.

Gibbs, C. S.: 1934. Preliminary studies on neurolymphomatosis and some more or less related diseases. Mass. Agr. Exper. Sta., Bul. 308.

——— and Johnson, C. G.: 1935. Leukosis and avian paralysis. Mass. Agr. Exper. Sta., Bul. 315.

——— and Johnson, C. G.: 1936. Differentiation of the pathological cell in neurolymphomatosis from lymphocytes of the blood of chickens. The differentiation of neurolymphomatosis from lympholeukosis. Mass. Agr. Exper. Sta., Bul. 327.

Gildow, E. M., Williams, J. K., and Lampman, C. E.: 1940. The transmission of and resistance to fowl paralysis (lymphomatosis). Ida. Exper. Sta., Bul. 235.

Glover, R. E.: 1940. Fowl paralysis. Transmission of infective agent to young chickens. Vet. Jour. 96:427.

Gohs, W.: 1934a. Über die Wirkung arteigener Knochen- und Knochenmarkzerfallstoffe auf die Knochen- und Blutbildung der Hühner. (Experimentell bei Hühnern erzeugte Osteody-strophia fibrosa, myeloische Leukose, Erythrämia und schwere Anämie.) Frankf. Zeitschr. f. Path. 46:453.

———: 1934b. Knochenveränderungen bei experimentell bei Hühnern erzeugter Osteodystrophia fibrosa. Frankf. Zeitschr. f. Path. 47:63.

Gray, E.: 1938. Pathogenic organisms and fowl paralysis. Vet. Record 50:1051.

———: 1940. Some experiments upon the therapeutic treatment of fowl paralysis (lymphomatosis) of poultry and the value of iodine in relieving the symptoms of such cases. Vet. Jour. 96:28.

Hall, W. J., Bean, C. W., and Pollard, M.: 1941. Transmission of fowl leukosis through chick embryos and young chicks. Am. Jour. Vet. Res. 2:272.

——— and Pollard, M.: 1943. Further studies on the propagation of fowl leucosis in chick embryos by intravenous inoculation. Am. Jour. Vet. Res. 4:287.

Hamilton, H. P.: 1934. Fowl paralysis, a disclaimer. Vet. Record 14:416.

Hamilton, C. M., and Sawyer, C. E.: 1939. Transmission of erythroleukosis in young chickens. Poultry Sci. 18:388.

Harriss, S. T.: 1939. Lymphomatosis (fowl paralysis) in the pheasant. Vet. Jour. 95:104.

———, Johnston, J. W., and Mitchell, S. G. A.: 1947. A study of isolation in fowl paralysis (lymphomatosis). Vet. Jour. 103:301.

Heisdorf, A. J., Brewer, N. R., and Lamoreux, W. F.: 1947. The genetic relationship between mortality from induced and spontaneous lymphomatosis. Poultry Sci. 26:67.

Hepding, L.: 1936. Beiträge zur Aetiologie und Diagnostik der ansteckenden Hühnerlähmung. Zeitschr. Infekt-Krankh. der Haustiere 49:292.

———: 1939. Ueber Toxoplasmen (Toxoplasma gallinarum n. sp.) in der Retina eines Huhnes und über deren Beziehung zur Hühnerlähmung. Zeitschr. Infekt-Krankh. der Haustiere 55:109.

Hutt, F. B.: 1932. Eight new mutations in the domestic fowl. Proc. Sixth Internat. Cong. Genetics 2:96.

———: 1939. Breeding strains of poultry resistant to fowl paralysis. Harper-Adams. Util. Poultry Jour. 24:395.

———: 1944. Simplified control of fowl leukosis. Farm Res. N. Y. St. and Cornell Sta. 10:11.

———: 1945. The high cost of fowl leucosis. Jour. Am. Vet. Med. Assn. 106:174.

——— and Cole, R. K.: 1947. The comparative importance of genes and of supposed egg-borne agents in the etiology of avian lymphomatosis. (Abst.) Poultry Sci. 26:544.

———, Cole, R. K., Ball, M., Bruckner, J. H., and Ball, R. F.: 1944. A relation between environment to two weeks of age and mortality from lymphomatosis in adult fowls. Poultry Sci. 23:396.

———, Cole, R. K., and Bruckner, J. H.: 1941. Four generations of fowls bred for resistance to neoplasms. Poultry Sci. 20:514.

Jaensch, P. A., and Lerche, G.: 1933. Die Augenveränderungen bei Marekscher Geflügellähme. Graefes Arch. f. Ophthalm. 131:359.

Jármai, K.: 1930. Beiträge zur Kenntnis der Hühnerleukose. Arch. f. wiss. u. prakt. Tierheilk. 62:113.

———: 1933. Infektionsversuche bebrüteter Eier mit dem "Virus" der Hühnererythroleukose. Deut. tierärztl. Wochenschr. 41:418.

———: 1934. Die Leukosen der Haustiere. Ergeb. der Allg. Path. Mensch. u. Tiere. 28:277.

———: 1935. Tumorerzeugung mit dem Leukoseagens der Hühner. Arch. f. wiss. u. prakt. Tierheilk. 69:275.

———: 1936. Zur Produktion und Artspezifizität des Agens der Hühnerleukose. Arch. f. wiss. u. prakt. Tierheilk. 70:62.

———: 1939. Über die Röntgenresistenz des Agens der übertragbaren Hühnerleukose im Vergleiche zu einigen übertragbaren Tiergeschwülsten und zu den Agenzien der übertragbaren Hühnersarkome. Arch. f. wiss. u. prakt. Tierheilk. 74:67.

———, Stenszky, T., and Farkas, L.: 1932. Neuere Beiträge zur Kenntnis der übertragbaren Hühnerleukose. Arch. f. wiss. u. prakt. Tierheilk. 65:46.

Johnson, E. P.: 1932. A study of lymphomatosis of fowls. Va. Agr. Exper. Sta., Tech. Bul. 41.

———: 1934. The etiology and histogenesis of leucosis and lymphomatosis of fowls. Va. Agr. Exper. Sta., Tech. Bul. 56.

———: 1937. Transmission of fowl leukosis. Poultry Sci. 16:255.

———: 1941. Fowl leukosis—manifestations, transmission, and etiological relationship of various forms. Va. Agr. Exper. Sta., Tech. Bul. 76.

———: 1945. Experimental vaccination for prevention of the avian leucosis complex. Am. Jour. Vet. Res. 6:198.

——— and Bell, W. B.: 1936. The blood pH of leukotic fowls and the filtrability of the leukosis agent. Jour. Infect. Dis. 58:342.

——— and Conner, B. V.: 1933. Blood studies of fowls with various forms of lymphomatosis (fowl paralysis). Jour. Am. Vet. Med. Assn. 83:325.

Jones, E. E.: 1934. Epidemic tremor, an encephalomyelitis affecting young chickens. Jour. Exper. Med. 59:781.

Jordan, H. E.: 1936. The relation of lymphoid tissue to the process of blood production in avian bone marrow. Am. Jour. Anat. 59:249.

——— and Johnson, E. P.: 1935. Erythrocyte production in the bone marrow of the pigeon. Am. Jour. Anat. 56:71.

Jungherr, E.: 1933. Observations on the macroscopic diagnosis of fowl paralysis. Poultry Sci. 12:184.

———: 1934. Studies on fowl paralysis. 1. Diagnosis. Storrs Agr. Exper. Sta., Bul. 200.

———: 1935. The etiologic and diagnostic aspects of the fowl paralysis problem. Jour. Am. Vet. Med. Assn. 86:421.

———: 1937. Studies on fowl paralysis. 2. Transmission Experiments. Storrs Agr. Exper. Sta., Bul. 218.

———: 1939. Neurolymphomatosis phasianorum. Jour. Am. Vet. Med. Assn. 94:49.

———: 1940. Wheat germ oil in the control of fowl paralysis. Poultry Sci. 19:94.

———, Durant, A. J., and Lee, C. D.: 1941. Tentative pathologic nomenclature for the disease complex variously designated as fowl leukemia, etc. Am. Jour. Vet. Res. 2:116.

——— and Landauer, W.: 1938. Studies on fowl paralysis. 3. A condition resembling osteopetrosis (Marble Bone) in common fowl. Storrs Agr. Exper. Sta., Bul. 222.

——— and Wolf, A.: 1939. Gliomas in animals. A report of two astrocytomas in the common fowl. Am. Jour. Cancer 37:493.

Kabat, E. A., and Furth, J.: 1941. Neutralization of the agent causing leukosis and sarcoma of fowls by rabbit antisera. Jour. Exper. Med. 74:257.

Kaupp, B. F.: 1921. Paralysis of the domestic fowl. Jour. Am. Assn. of Instructors and Invest. in Poult. Husb. 7:25.

Kennard, D. C., and Chamberlin, V. D.: 1934. Poultry mortality. Ohio Agr. Exper. Sta., Bimo. Bul. 19:137—42. (Bul. 169.)

Kissling, R. E.: 1947. Leukoagglutination as a serological diagnosis for avian lymphomatosis. Poultry Sci. 26:74.

Kitt, T.: 1931. Die Leukomyelose der Hühner. Ergeb. der Hyg., Bakt. Immunitätsforsch. u. exper. Therap. 12:15.

Ledingham, J. C. G., and Gye, W. E.: 1935. On the nature of the filterable tumour-exciting agent in avian sarcomata. Lancet 1:376.

Lee, C. D.: 1942. Studies on production of specific antibodies against the agent of fowl leucosis. Am. Jour. Vet. Res. 3:336.

——— and Wilcke, H. L.: 1941. Transmission experiments with iritis of fowls. Am. Jour. Vet. Res. 2:292.

Lerche, M., and Fritzsche, K.: 1934. Histopathologie und Diagnostik der Geflügellähme. Zeitschr. f. Infekt-Krankh. d. Haustiere 45:89.

Magnusson, H.: 1935. Hönsparalysi. Svensk. Vet. Tidskr. 40:43.

Magnusson, H.: 1946. Om en ovanlig skelettsjukdom hörande till hönsleukoskomplexet. Skand. Veterinärtidskrift för bakr., patol. samt kött-och mjölkhyg. 36:70.

Marek, J.: 1907. Multiple Nervenentzündung (Polyneuritis) bei Hühnern. Deutsch. tierärztl. Wochenschr. 15:417.

Marine, D., and Rosen, S. H.: 1941. Sex hormones and lymphomatosis in fowls. Proc. Soc. Exper. Biol. and Med. 47:61.

Mathews, F. P.: 1929. Leukochloroma in the common fowl. Its relation to myelogenic leukemia and its analogies to chloroma in man. Arch. Path. 7:442.

———— and Walkey, F. L.: 1929. Lymphadenomas of the common fowl. Jour. Canc. Res. 13:383.

McClary, C. F., and Upp, C. W.: 1939. Is paralysis of fowls, as manifested by iritis, transmitted through the egg? Poultry Sci. 18:210.

McGaughey, C. A., and Downie, A. W.: 1930. Preliminary report on an outbreak of fowl paralysis in England. Jour. Comp. Path. and Therap. 43:63.

McIntosh, J.: 1933. On the nature of tumors induced in fowls by injections of tar. Brit. Jour. Exper. Path. 14:422.

———— and Selbie, F. R.: 1939. Further observations on filtrable tumors induced in fowls by injections of tar. Brit. Jour. Exper. Path. 20:49.

McLennan, G. C.: 1935. "Range paralysis": *Neuro-lymphomatosis gallinarum*. Australian Vet. Jour. 11:42.

Moynihan, I. W.: 1943. Avian osteopetrosis. Canad. Jour. Comp. Med. and Vet. Sci. 7:327.

Murphy, J. B., and Sturm, E.: 1941a. Further investigation of induced tumors in fowls. Cancer Res. 1:477.

———— and Sturm, E.: 1941b. Further investigation on the transmission of induced tumors in fowls. Cancer Res. 1:609.

Nelson, N. M., and Thorp, Jr., F.: 1943. Ocular lymphomatosis with special reference to chromatism of irides. Am. Jour. Vet. Res. 4:294.

Norris, L. C., Caskey, C. D., and Bauernfeind, J. C.: 1940. Malformation of the tarso-metatarsal and phalangeal bones of chicks. Poultry Sci. 19:219.

Nyfeldt, A.: 1934. Étude sur les leucosis des poules. I. Une myéloblastose pure. Sang. 8:566.

Oakley, C. L.: 1935. Lymphomatosis. Proc. Roy. Soc. Med. 28:999.

Oberling, Ch., and Guérin, M.: 1933. Lésions tumorales en rapport avec la leucémie transmissible des poules. Bul. Assoc. franc. pour Étude Canc. 22:180.

———— and Guérin, M.: 1933a. Nouvelles recherches sur la production de tumeurs malignes avec le virus de la leucémie transmissibles des poules. Bul. Assoc. franc. pour Étude Canc. 22:326.

———— and Guérin, M.: 1933b. Nouvelles recherches sur les ostéites par carence chez les poules. Compt. rend. Soc. de biol. 112:1288.

———— and Guérin, M.: 1934. La leucémie érythroblastique ou érythroblastose transmissible des poules. Bul. Assoc. franc. pour Étude Canc. 23:38.

————, Guérin, M., and Boic, V.: 1933. Recherches sur la leucémie transmissible (érythroblastose) des poules. Compt. rend. Soc. de biol. 112:559.

———— and Muller, J.: 1934. Tentatives d'homogreffes parathyroidiennes chez des poules carencées par un régime pauvre en calcium. Ann. Anat. Path. et Anat. Norm. Méd.-Chir. 11:744.

Olson, Jr., C.: 1936. A study of transmissible fowl leukosis. Jour. Am. Vet. Med. Assn. 89:681.

————: 1937. Attempts to transmit fowl paralysis. Jour. Infect. Dis. 61:325.

————: 1940. Transmissible fowl leukosis. A review of the literature. Mass. Agr. Exper. Sta., Bul. 370.

————: 1941. A transmissible lymphoid tumor of the chicken. Cancer Res. 1:384.

————: 1945a. The immunizing action of a lymphoid tumor in chickens. Am. Jour. Vet. Res. 6:103.

————: 1945b. Immunization against a lymphoid tumor of the chicken. I. Attenuation by freezing. Cornell Vet. 35:221.

————: 1947. Immunization against a lymphoid tumor of the chicken. IV. Use of miscellaneous tissues. Cornell Vet. 37:231.

———— and Zeissig, A.: 1936. A study of the antigenic properties of certain normal and pathological lymphoid deposits in tissues of chickens. Jour. Immunol. 31:309.

Pappenheimer, A. M., Dunn, L. C., and Cone, V.: 1926. A study of fowl paralysis (*neuro-lymphomatosis gallinarum*). Storrs Agr. Exper. Sta., Bul. 143.

————, Dunn, L. C., and Cone, V.: 1929a. Studies on fowl paralysis (*neuro-lymphomatosis gallinarum*). I. Clinical features and pathology. Jour. Exper. Med. 49:63.

————, Dunn, L. C., and Seidlin, S. M.: 1929b. Studies on fowl paralysis (*neuro-lymphomatosis gallinarum*). II. Transmission experiments. Jour. Exper. Med. 49:87.

Patterson, F. D.: 1934. *Neurolymphomatosis gallinarum*. Proc. Twelfth Internat. Vet. Cong. (New York) 3:265.

————: 1936. Fowl leukosis. Jour. Am. Vet. Med. Assn. 88:32.

————, Wilcke, H. L., Murray, Chas., and Henderson, E. W.: 1932. So-called range paralysis of the chicken. Jour. Am. Vet. Med. Assn. 81:747.

Pentimalli, F.: 1915. Über die Geschwülste bei Hühnern. I. Mitteilung. Allgemeine Morphologie der spontanen und der transplantablen Hühnergeschwülste. Zeitschr. f. Krebsforsch. 15:111.

Pentimalli, F.: 1941. Transplantable lymphosarcoma of the chicken. (Abst.) Cancer Res. 1:69.

Phillips, P. H., and Engel, R. W.: 1938. The histopathology of neuromalacia and "curled toe" paralysis in the chick fed low riboflavin diets. Jour. Nutr. 16:451.

Pikovski, M., and Doljanski, L.: 1946. Bone tumors in fowls injected intravenously with causative agent of Rous sarcoma. Proc. Soc. Exper. Biol. and Med. 61:246.

Pollard, M., and Hall, W. J.: 1911. Preliminary report on inter-species transmission of avian leucosis in embryos. Jour. Am. Vet. Med. Assn. 99:218 '(Abst.).

———, Hall, W. J., and Eichhorn, A.: 1943. Serological studies on the avian leukosis complex. Preliminary report on complement fixation. Poultry Sci. 22:20.

Potel, K.: 1938. Hyalin-schollige Degeneration der Skeletmuskulature bei Kücken und Junghühnern. Arch. f. wiss. u. prakt. Tierheilk. 72:216.

Pugh, L. P.: 1927. Sporadic diffuse osteo-periostitis of fowls. Vet. Record 7:189.

Reinhardt, R.: 1930. Die pathologisch-anatomischen Veränderungen bei den spontanen Krankheiten der Hausvögel. Ergeb. d. allgem. Path. u. path. Anat. des Mensch. u. der Tiere. 23:553 (p. 682).

Reis, J., and Nobrega, P. (com. collab. Reis, A. S.): 1936. Doencas das Aves. (Tratado de ornithopathologia.) Instituto Biologico, São Paulo, Brazil, p. 417.

Ringoen, A. R.: 1934. The inter-sinusoidal capillaries of avian bone marrow. Anat. Record 58:84 (Suppl.).

Rothe Meyer, A., and Engelbreth-Holm, J.: 1933. Experimentelle Studien über die Beziehungen zwischen Hühnerleukose und Sarkom an der Hand eines Stammes von übertragbarer Leukose-Sarkom Kombination. Acta. Path. et Microbiol. Scand. 10:380.

Sanders, E., Huddleson, I. F., and Schaible, P. J.: 1944. An electrophoretic study of serum and plasma from normal and leucosis-affected chickens. Jour. Biol. Chem. 155:469.

Schmeisser, H. C.: 1915. Spontaneous and experimental leukemia of the fowl. Jour. Exper. Med. 22:820.

Seagar, E. A.: 1933. Cellular reactions in the blood in *neuro-lymphomatosis gallinarum* (fowl paralysis). Vet. Jour. 89:557.

Stern, G., and Kirschbaum, A.: 1939. On the nature of the agent causing leucosis in fowls. Science 89:610.

Storti, E., and Mezzadra, G.: 1938. Tentatives de culture du virus de la leucémie des poules dans la membrane chorio-allantoide. (Note préliminaire.) Sang. 12:533.

Stubbs, E. L.: 1938. Fowl leukosis. Jour. Am. Vet. Med. Assn. 92:73.

——— and Furth, J.: 1932. Anemia and erythroleucosis occurring spontaneously in the common fowl. Jour. Am. Vet. Med. Assn. 81:209.

——— and Furth, J.: 1935. The relation of leukosis to sarcoma of chickens. I. Sarcoma and erythroleukosis (strain 13). Jour. Exper. Med. 61:593.

Taylor, L. W., and DeOme, K. B.: 1939. Failure of wheat germ oil to prevent lymphomatosis in chickens. Jour. Am. Vet. Med. Assn. 95:73.

———, Lerner, I. M., DeOme, K. B., and Beach, J. R.: 1943. Eight years of progeny-test selection for resistance and susceptibility to lymphomatosis. Poultry Sci. 22:339.

Thiersch, J. B.: 1944. Attempts to transmit leucaemia of man and of mice to the chick embryo and to the young chick by the amniotic and intravenous routes. Australian Jour. Exper. Biol. and Med. Sci. 22:57.

Thomsen, O., and Engelbreth-Holm, J.: 1932. De la provocation expérimentale d'états leucosiformes chez les poules. Compt. rend. Soc. de biol. 109:1213.

Troisier, J., with the collab. of Sifferlen, J.: 1935. Leucose et sarcomatose des poules, unité de virus. Ann. d'Inst. Past. 55:501.

Uhl, E.: 1938. Active immunization of chickens against chicken leukosis with agent adsorbed by aluminum hydroxide. Acta Path. et Microbiol. Scand. Suppl. 37:544.

Upp, C., and Tower, B. A.: 1936. The incidence of blindness and paralysis according to family. Poultry Sci. 15:421.

Van den Berghe, L., and d'Ursel, Fr.: 1939. Erythroblastose du poussin éclos après inoculation chorio-allantoidienne du virus (souche O.G.). Compt. rend. Soc. de biol. 131:1302.

Van der Walle, N., and Winkler-Junius, E.: 1924. De Neuritis-Epizootie bij Kippen te Barneveld in 1921. Tijdschr. voor verg. Geneesk. 10:34.

Venkataraman, R.: 1936. A note on osteitis deformans in two fowls. Indian Jour. Vet. Sci. 6:108.

Wagener, K.: 1939. Untersuchungen über die Übertragbarkeit der Geflügellähme. Proc. Seventh World's Poultry Cong.:301.

Warrack, G. H., and Dalling, T.: 1932. So-called fowl paralysis Also called neuritis in chickens, range paralysis, *neurolymphomatosis gallinarum.* (A discussion on the various theories as to causation with special reference to field observations and laboratory transmission experiments.) Vet. Jour. 88:28.

Waters, N. F.: 1945a. Natural transmission of avian lymphomatosis. Poultry Sci. 24:226.

———: 1945b. Breeding for resistance and susceptibility to avian lymphomatosis. Poultry Sci. 24:259.

———: 1947. The contagious nature of a lymphoid tumor in chickens. Science 106:246.

———: 1945c. Lymphomatosis in chickens as influenced by diallel crossing. Poultry Sci. 24:387.

Waters, N. F., and Prickett, C. O.: 1944. The development of families free of lymphomatosis. Poultry Sci. 23:321.

Wickware, A. B.: 1943. Transmissible leucosis. A recently isolated strain. Canad. Jour. Comp. Med. 7:265.

———: 1946. The incidence of erythroleucosis following inoculation by various routes. Canad. Jour. Comp. Med. 10:74.

Wilcke, H. L., Patterson, F. D., Henderson, E. W., and Murray, C.: 1933. The effect of the ration upon the incidence of so-called range paralysis. Poultry Sci. 12:226.

## CHAPTER NINETEEN

## INFECTIOUS BRONCHITIS

*By* J. R. BEACH, *Department of Veterinary Science,*
*University of California, Berkeley, California*

☆   ☆   ☆

This disease was first studied and described by Schalk and Hawn (1931) in North Dakota under the title, "An apparently new respiratory disease of baby chicks." They reported having received information that what appeared to be an identical disease was being encountered in Mississippi, Louisiana, Ohio, Illinois, Nebraska, Iowa, and South Dakota. By 1933 it had been recognized in California (Beach, 1933, 1934), Kansas (Bushnell and Brandly, 1933), and possibly also in Massachusetts (Gibbs, 1933). Its distribution was soon found to be nationwide. It is probable that it was not identified earlier in some sections because it was mistaken for laryngotracheitis, which it closely resembles. The disease became known popularly as gasping disease or chick bronchitis. However, the scientific name, infectious bronchitis, as was suggested by Schalk and Hawn (1931), has been accepted and has come into common usage. Originally, the disease was believed to be confined to young chicks, but in some sections of the country it has become of greater importance as a disease of partly grown and laying pullets.

**Etiology.** Infectious bronchitis is caused by a filtrable virus which readily passes through all grades of Berkefeld filters (Beach and Schalm, 1936). The virus is found most abundantly in the exudate and tissues of the respiratory organs affected. Bushnell and Brandly (1933), however, reported successful transmission of the disease with blood, spleen, liver, and kidney tissues of affected chicks. In exudate dried after freezing and stored in a refrigerator, the virus has remained viable for 180 days; in 50 per cent glycerin and stored in a refrigerator, it remained viable for 80 days (Beach and Schalm, 1936). There is little definite information concerning the time the virus will remain active under natural conditions. In many instances inability to transmit the disease by the injection of exudate from dead birds suggests that the virus perishes quickly after the death of the host. On the other hand, the conditions under which some natural outbreaks have occurred give the impression that the virus may remain active in a contaminated brooder house or on equipment for several months. Field evidence suggests that recovered birds

may become healthy carriers of the virus and thus perpetuate the disease on a farm from year to year. Hofstad (1947) attempted to demonstrate this experimentally by contact exposure of susceptible birds to recovered ones. He failed to show by this procedure, however, that the carrier state would persist in chickens longer than 35 days after recovery.

Beaudette and Hudson (1937) found that the virus can be propagated in chicken embryos. Little observable effect of the virus on the embryos was noted during the first passages. The virus rapidly became adapted to embryos, however, and after a few passages death of the embryos became a regular occurrence. Delaplane and Stuart (1939, 1941) reported similar results from embryo-culture of the virus and also that continued cultivation of the virus in embryos was accompanied by progressive decrease in the virulence of the virus for chickens. Later Delaplane (1947) found that more rapid adaptation of the virus to embryos occurred when inoculation was into the allantoic cavity instead of on the chorio-allantoic membrane as had previously been the custom. This was evidenced by a dwarfing effect on the embryo which was observable at the first passage and became more distinct at the second and third passages. The behavior of the virus in embryos is considered sufficiently characteristic and different from that of other viruses which affect chickens that the embryo-culture technique can be utilized in differentiating infectious bronchitis from clinically similar diseases such as infectious laryngotracheitis and pneumoencephalitis (Newcastle disease).

**Symptoms.** Gasping is the most characteristic and predominant symptom. When a chick is severely affected, the beak is pointed upward and opened wide at each inspiration. Convulsive coughing is also seen. Chicks less severely affected exhibit such symptoms irregularly, but if one is held close to the ear, short, crackling râles may be heard. Nasal discharge and swollen sinuses are frequently seen and may be predominant, especially in chicks a few weeks old. Beach and Schalm (1936) reported that nasal passage involvement occurred in more than half of the chicks artificially infected by the intratracheal route. Depression and weakness are seen in the advanced stage of the disease.

In chicks the disease occurs most frequently when the birds are under three or four weeks of age, and it has been reported in birds as young as 2 days. Beach and Schalm (1936) found that chicks from 10 to 112 days old were equally susceptible to artificial infection. The disease spreads rapidly and is apt to infect nearly all of a flock within a brief period. In young chicks the disease may vary from one of a mild, relatively harmless, transitory nature to a more damaging type causing 25 to 90 per cent mortality. The effect on birds from the age of six weeks up to shortly before the first egg is laid is not likely to be more than slight retardation in their development. Infection in a laying flock usually causes serious economic loss from depression of the egg yield, but seldom is the loss from death of serious consequence.

Schalk and Hawn (1931) state that the most constant and characteristic lesion is marked congestion of the lungs, usually distributed through both lobes, and the bronchi and bronchioles may be entirely or partially filled with sero-mucoid liquid exudate. Others have reported that the presence of clear, turbid, or thick yellow mucous exudate or caseous plugs in the lower trachea and bronchi is the most characteristic autopsy finding. The same sort of exudate is found in the nasal chambers when they are involved. An accumulation of caseous or thick, viscid mucus is occasionally seen in the larynx and air sacs.

Affected laying birds show gasping and coughing much like those with laryngotracheitis. Symptoms of nasal involvement have not been reported. Spread of the disease through a flock is very rapid and the morbidity is high, but the mortality is negligible or absent. Considerable economic loss is sustained, however, from the sharp drop in egg production which invariably occurs and may persist for several weeks.

At autopsy, varying amounts of clear or turbid, liquid or viscid exudate are found in the trachea, and only rarely is it of the thick, yellowish, and bloody character of that found in laryngotracheitis.

The disease in semimature and laying chickens appears to have occurred with variable frequency in different localities. Schalk and Hawn (1931) state, "Of the scores of farms suffering from the ravages of this disease in baby chicks, only two reported respiratory disturbances in older fowls." Beaudette and Hudson (1937) note that "usually it occurs in young chicks, but occasionally the disease is spread to the adults on the same farm." The writer has consistently failed to establish the presence of the infectious bronchitis virus in outbreaks of bronchitis-like disease among flocks of maturing and laying pullets in districts in which the disease was prevalent among chicks. On the other hand, information from several sources indicates that, in certain states, the disease is more prevalent in semimature and laying pullets than in brooder chicks.

**Transmission.** The disease spreads readily from affected to healthy fowls by either direct or indirect contact. It is usually easily transmitted by the intranasal or intratracheal injection of exudate. Transmission by subcutaneous, intramuscular, or intraperitoneal injection, or by swabbing the cloacal mucous membrane with exudate has also been reported. Levine and Hofstad (1947) demonstrated experimentally by specially designed apparatus, that the infection can be air-borne for a distance of at least 5 feet. The source of the first infection on a farm is often obscure. Numerous outbreaks, however, have been traceable to hatcheries which deal in "started chicks" and in which the disease has become established.

**Diagnosis.** It is difficult to identify infectious bronchitis by clinical manifestations and autopsy findings because they are so similar to those of other prevalent respiratory diseases. In case coughing and gasping are the only

symptoms present in an affected flock of chicks, the disease is likely to be infectious bronchitis but could be pneumoencephalitis. On the other hand, if some of the affected chicks develop symptoms of central nervous system involvement, pneumoencephalitis would be the more probable diagnosis. In older birds, infectious bronchitis must be differentiated from laryngotracheitis and pneumoencephalitis (Chapters 20 and 21), and when there is nasal involvement, from infectious coryza (Chapter 13).

Laboratory procedures are often required for definite identification of the disease present. The various ones which may be employed are transmission trials, serum-virus neutralization, and hemagglutination-inhibition tests (see pages 501–508 in Chapter 21 for a description of these tests), and isolation of the virus in embryo culture. Delaplane (1947) described a technique for isolation of virus in embryo culture which he considers particularly suited for the differentiation of infectious bronchitis and pneumoencephalitis. (See page 501.)

**Prevention.** The only recommendation for prevention that can be given at present is the strict application of the measures of hygiene and sanitation. It is particularly important to be certain that all new stock comes from a clean source and that houses and equipment be thoroughly cleaned and disinfected after an outbreak. Contact between survivors of an outbreak and susceptible birds should be avoided. Depopulation may be necessary to eradicate the disease from an infected broiler plant or other establishment at which continuous brooding is practiced. Recovered birds are immune, but no safe method of artificial immunization has been found. Cloacal vaccination, as used for laryngotracheitis, is not applicable because infection of the respiratory organs can be produced by applying virus to the cloaca.

An immunization procedure to prevent the occurrence of infectious bronchitis among laying flocks has been used to a limited extent in Massachusetts since 1941, and more recently has been started in other New England states. According to information received in personal communications from Henry Van Roekel, Massachusetts Agricultural Experiment Station, and E. F. Waller, Vermont State Agricultural College, the essential features of the immunizing procedure is as follows: A few chicks of a flock of suitable age are inoculated with the virus and from these the infection spreads throughout the remainder of the chicks. The inoculum is either tracheal exudate from artificially infected chicks or virus which has been propagated in embryos. It is supplied by the Agricultural Experiment Stations, and is administered by trained field men. Distribution of the virus is restricted to farms on which previous occurrence of the disease has been definitely determined by laboratory diagnosis. The chicks are inoculated when of an age at which the disease is least harmful, i.e., not less than six or eight weeks nor more than sixteen weeks old. The results in general are said to have been satisfactory

and especially so when the birds were ten to fourteen weeks of age, in good health, and inoculated during the summer months.

Levine and Hofstad (1947) reported that infectious bronchitis can be destroyed by exposure to ultraviolet radiation under certain experimental conditions but concluded that under practical field conditions, "lamps emitting ultraviolet light will have little value in the control of respiratory diseases in fowls."

**Treatment.** No effective treatment for either individuals or a flock has been found.

## REFERENCES

Beach, J. R.: 1933. Poultry disease; recent discoveries. Proc. 5th Pacific Sci. Cong. 4:2961.

———: 1934. Coryza and other respiratory infections in chickens. Proc. 12th Internat. Vet. Cong. 3:144.

——— and Schalm, O. W.: 1936. A filtrable virus distinct from that of laryngotracheitis, the cause of a respiratory disease of chicks. Poultry Sci. 15:199.

Beaudette, F. R., and Hudson, C. B.: 1937. Cultivation of the virus of infectious bronchitis. Jour. Am. Vet. Med. Assn. 90:51.

Bushnell, L. D., and Brandly, C. A.: 1933. Laryngotracheitis in chicks. Poultry Sci. 12:55.

Delaplane, J. P.: 1947. Technique for the isolation of infectious bronchitis or Newcastle virus including observations on the use of streptomycin in overcoming bacterial contaminants. Mimeo. report.

——— and Stuart, H. O.: 1939. Studies of infectious bronchitis. R. I. Agr. Exper. Sta., Bul. 273.

——— and Stuart, H. O.: 1941. The modification of infectious bronchitis virus of chickens as a result of propagation in embryonated chicken eggs. R. I. Agr. Exper. Sta., Bul. 284.

Gibbs, C. S.: 1933. Bronchitis of baby chicks. Poultry Sci. 12:46.

Hofstad, M. S.: 1947. A study of infectious bronchitis in chickens. Cornell Vet. 37:29.

Levine, P. P., and Hofstad, M. S.: 1947. Attempts to control air-borne infectious bronchitis and Newcastle disease of fowls with sterilamps. Cornell Vet. 37:204.

Schalk, A. F., and Hawn, M. C.: 1931. An apparently new respiratory disease of baby chicks. Jour. Am. Vet. Med. Assn. 78:413.

# CHAPTER TWENTY

# INFECTIOUS LARYNGOTRACHEITIS

*By* J. R. Beach, *Department of Veterinary Science,*
*University of California, Berkeley, California*

☆ ☆ ☆

In the early reports concerning this disease it was termed infectious bronchitis (Beach, 1925; Kernohan, 1930; Beaudette and Hudson, 1930) or tracheo-laryngitis (May and Tittsler, 1925). Later, however, the histopathological studies of the disease by Seifried (1931) showed that it affected principally the larynx and trachea and in that order, and the more correct term, infectious laryngotracheitis, was therefore adopted. Since its first recognition in 1924 as a distinct disease of chickens, the disease has assumed a position of major economic importance to the poultry industry of the United States and Canada. It was positively identified in Australia (Seddon and Hart, 1935, 1936) in 1935 and subsequently has been observed in several European countries.

**Etiology.** The cause of the disease was definitely established to be a filtrable virus in 1930 (Beach, 1930, 1931a, 1931b). The virus is found in abundance in the exudate present in the trachea of infected birds. It has also been demonstrated in livers and spleens of some chickens. Its presence there is believed to be due to accidental entrance of the virus into the blood stream through injured walls of blood vessels of the larynx and does not imply any real involvement of organs (Beach, 1931a).

It has been shown experimentally that the virus is quickly destroyed by exposure to moderate temperature (55° to 75° C.), does not survive longer than 90 days at room temperature, may be killed by exposure to direct sunlight in 7 hours, is readily destroyed by a 3 per cent solution of cresol disinfectant and a 1 per cent solution of sodium hydrate (lye), and survives in the body of a dead chicken only until decomposition begins (Schalm and Beach, 1935). These findings indicate that the virus is not likely to survive on a farm from one season to another outside of a living host and that saponated cresol and lye are efficient disinfectants for use on buildings and equipment after an outbreak.

**Symptoms.** The predominant symptoms are gasping and coughing. The affected fowl assumes a sitting position, and when severely affected, the neck is drawn in, the beak is pointed downward, and eyes are closed (Fig. 20.1).

At each inhalation the head is thrust forward and upward with the beak open, and the intake of air may be accompanied by a loud wheezing sound (Fig. 20.2). Spasmodic exhalation or coughing is frequent and often results in the expulsion from the trachea of a mass of mucus or clotted blood. In less severe cases, gurgling or rattling during breathing is the only manifestation observed. Examination may reveal the larynx to be nearly filled with a bloody or yellow thick mucus, or with caseous material, while in other cases, in a live



FIG. 20.1. An advanced case of laryngotracheitis. Attitude during expiration. (Beach and Freeborn, Univ. of Calif.)

bird, the cause of the respiratory distress is not visible. Rarely the disease may involve also the eyes and nasal passages.

The disease usually has a sudden onset and spreads very rapidly through a flock. The morbidity is very high, but the mortality ranges from 5 to 60 per cent or higher, the average being about 15 per cent during a period of from two to four weeks (Hinshaw, Jones, and Graybill, 1931; Hinshaw, 1931). Occasionally, the spread is slow, and evidence of the disease may exist in a flock for weeks. The production of laying flocks is greatly reduced or ceases entirely, and does not again reach the normal rate for one or two months. The value of eggs lost from decreased production often exceeds that of the birds which die. Fowls which survive 2 or 3 days of illness are likely to make prompt and complete recovery. After recovery, however, a bird may carry the virus for an indefinite period (Gibbs, 1932; Hudson and Beaudette, 1932a). Such healthy carriers are the principal means of perpetuating the disease on a farm from year to year, and may also be the means of its first introduction on a farm.

The lesions found at autopsy are confined to the larynx and trachea; the linings of these organs are inflamed and their lumens wholly or partially filled with mucus mixed with varying amounts of clotted blood. In cases of 2 or 3 days duration the mucous exudate becomes caseated. Occasionally, a hollow cast of caseous mucus is found extending the entire length of the trachea. In some cases the material present appears to consist entirely of clotted blood. The collection of exudate, in some cases, is confined to the



FIG. 20.2. Laryngotracheitis. Same fowl as Figure 20.1. Attitude during inspiration.

lower portion of the trachea, and in others to the upper trachea and larynx. Death is due entirely to asphyxiation which occurs when the lumen of the larynx becomes filled with a mass of mucous or caseous exudate or clotted blood which the birds are unable to expel by their convulsive coughing. The lungs usually appear normal except for small areas of congestion. Other organs are not involved.

**Host specificity of the disease.** Laryngotracheitis has occurred or been produced only in chickens and pheasants. Domesticated ducks, turkeys, pigeons, and wild and free-flying species of birds, including sparrows, crows, starlings, doves, and quail have been found refractory, and so, too, have

rabbits, guinea pigs, and white rats (Beach, 1931b; Hudson and Beaudette, 1932b).

**Diagnosis.** Many outbreaks of laryngotracheitis can be readily recognized by the characteristic symptoms, autopsy findings, and the rapidity of spread through a flock. Almost identical characteristics, however, are presented by infectious bronchitis and pneumoencephalitis (Newcastle disease). Respiratory disease of this character in young chicks is more likely to be infectious bronchitis or pneumoencephalitis, because laryngotracheitis is rarely seen in such young birds. A respiratory disease of older birds which has symptoms of laryngotracheitis but which is accompanied by few or no deaths and the collection of clear or slightly turbid mucus instead of yellowish or bloody mucus in the trachea is likely to be infectious bronchitis or pneumoencephalitis. In many cases, however, a positive diagnosis must be based upon the results of transmission and cross-protection tests.

In some outbreaks of a severe type of coryza, many of the affected fowls have tracheal involvement, and coughing and gasping are prominent symptoms. The diagnostic question in such cases is to determine whether the fowls have coryza alone or are affected with both coryza and laryngotracheitis. Transmission and cross-protection tests are usually necessary to provide the answer.

**Vaccination.** This procedure is based upon the discovery that the mucous membrane of the cloaca and bursa of Fabricius is susceptible to the virus of laryngotracheitis, that infection of these parts does not cause a systemic disturbance or spread to the respiratory tract, and that the cloacal infection subsides in about 7 days and the fowl is thereafter immune to infection of the respiratory tract (Hudson and Beaudette, 1932a). The practicability, effectiveness, and safety of the application of these findings to the vaccination of chickens on farms were demonstrated by extensive field trials by Beaudette and Hudson (1933); Beach, Schalm, and Lubbehusen (1934); and Gibbs (1933, 1934).

The vaccine originally consisted of tracheal exudate taken from artificially infected chickens. Since the demonstration by Burnet (1934), however, that the laryngotracheitis virus can be propagated in chicken embryos, and the extensive experimental studies of vaccination with embryo-propagated virus by Brandly (1935, 1936), and Beaudette and Hudson (1939), the embryo-propagated virus has been almost exclusively used for vaccine preparation. The advantage of vaccine of embryo origin over that of chicken origin is that the former can be produced bacteriologically sterile and cannot carry any other disease-producing agent which might be present in chickens used in the production of tracheal exudate. The virus-bearing exudate or embryo tissue, after removal, is dried and finely powdered. For use in vaccination the powdered virus is suspended in a solution of glycerin.

The vaccination procedure consists in brushing the vaccine onto the cloacal mucous membrane. The cloacal lips are forced open to expose the membrane which is then brushed until redness or even bleeding is produced. Another, though little used, method of vaccinating growing chickens consists of injection of the vaccine directly into the bursa of Fabricius through a blunt, slightly curved hypodermic needle (Beach, Schalm, and Lubbehusen, 1934). The resultant infection of the mucous membrane or "take" consists of swelling of the cloacal lips, redness and swelling of the mucous membrane (Fig. 20.3), and the presence of mucus, often yellow and



Fig. 20.3. Taken from laryngotracheitis vaccination: A—swelling of cloacal lips. B—reddening of mucous membrane. (Beach and Freeborn, Univ. of Calif.)

flecked with blood on its surface. A "take" usually reaches maximum intensity on the fifth day, and by the seventh day has nearly disappeared. In contrast to survivors of respiratory infection, the virus does not persist in the cloaca or bursa of Fabricius after the reaction subsides (Beach, 1935). The birds should be examined on the fifth day after vaccination, and any that do not show a definite "take" should be revaccinated immediately. If not revaccinated, the birds without a "take" can be expected to acquire severe respiratory infection from the virus eliminated by the birds with "takes." No additional special after-care is required. Chickens can be vaccinated at any time after they reach the age of six weeks, but preferably before they have begun to lay.

*Vaccination is indicated:*

1. For the prevention of the spread of the disease on a farm after it has appeared in one pen. In such cases the noninfected pens should be treated first.

2. For young stock on farms where the disease has occurred in the past and on which survivors of a past outbreak still remain.

3. For susceptible fowls which are added to a flock in which the disease is or has been present; or for a susceptible flock to which known survivors of the disease are to be added.

4. For healthy flocks, either already existing or newly established in a congested poultry district in which the disease is prevalent.

*Vaccination is contra-indicated:*

1. As a preventive measure in a flock not previously infected unless it is located in a congested poultry district in which the disease is prevalent.

2. For the control of an outbreak of any respiratory disease not definitely diagnosed as laryngotracheitis.

3. For a portion of the susceptible birds on a farm, unless the birds not vaccinated are too young, are well segregated, and are to be vaccinated as soon as they reach suitable age.

4. By persons unfamiliar with the hazards attendant upon the use of a virulent live-virus vaccine.

**Prevention.** The strict application of the principles of hygiene and sanitation should be adequate to prevent the introduction of the disease into a flock not located in a poultry district. Particular attention should be given to the source of added stock, and any article that is used in poultry houses.

Prevention of recurrence of the disease on a farm after an outbreak without annual vaccination may be accomplished by the following procedure:

1. All birds which have had or have been exposed to infectious laryngotracheitis are removed from the premises.

2. All buildings and equipment, including outer wearing apparel of attendants, used in the housing and care of the condemned chickens are thoroughly cleaned and disinfected.

3. The houses and equipment are left unused for at least two months after being cleaned and disinfected.

4. Chicks on the premises may be kept for restocking provided they are well separated from the condemned birds, and they are definitely known to have escaped infection.

5. New stock should come from absolutely clean sources, and preferably as baby chicks.

These measures would be of questionable effectiveness in poultry districts in which infectious laryngotracheitis is prevalent.

**Treatment.** No medicinal treatment for individual birds or a flock has been found to have sufficient merit to warrant its use. In fact dropping chemicals into the trachea or spraying them over fowls on the roost may increase the respiratory distress of affected birds. In cases of severe dyspnea due to occlusion of the larynx and upper trachea, quick relief and recovery can sometimes be obtained by careful removal of the material with forceps. This is the only type of treatment that has been found worth-while.

**REFERENCES**

Beach, J. R.: 1925. Infectious bronchitis of fowls. Jour. Am. Vet. Med. Assn. 68:570.
———: 1930. The virus of laryngotracheitis of fowls. Science 72:633.

————: 1931a. A bacteriological study of infectious laryngotracheitis of chickens. Jour. Exper. Med. 54:801.

————: 1931b. A filtrable virus the cause of infectious laryngotracheitis of chickens. Jour. Exper. Med. 54:809.

————: 1935. The survival of the virus of infectious laryngotracheitis in the bursa of Fabricius and cloaca of chickens after "intra-bursal" injections. Jour. Infect. Dis. 57:133.

————, Schalm, O. W., and Lubbehusen, R. E.: 1934. Immunization against infectious laryngotracheitis of chickens by "intrabursal" injection of virus. Poultry Sci. 13:218.

Beaudette, F. R.: 1939. The viability and immunizing value of egg-propagated laryngo-tracheitis virus. Jour. Am. Vet. Med. Assn. 95:333.

———— and Hudson, C. B.: 1930. Some observations on an outbreak of infectious bronchitis in battery brooded chicks. Proc. 22nd Ann. Meet. Poultry Sci. Assn., pp. 71–77. (Publ. as suppl. to Poultry Sci. 9, 1930.)

———— and Hudson, C. B.: 1933. Experiments on immunization against laryngotracheitis in fowls. Jour. Am. Vet. Med. Assn. 82:460.

Brandly, C. A.: 1935. Some studies of infectious laryngotracheitis. The continued propagation of the virus upon the chorio-allantoic membrane of the hen's egg. Jour. Infect. Dis. 57:201.

————: 1936. Studies on the egg-propagated viruses of infectious laryngotracheitis and fowl pox. Jour. Am. Vet. Med. Assn. 88:587.

Burnet, F. M.: 1934. The propagation of the virus of infectious laryngotracheitis on the chorio-allantoic membrane of the developing egg. Brit. Jour. Exper. Path. 15:52.

Gibbs, C. S.: 1932. Chronic carriers of infectious laryngotracheitis. Jour. Am. Vet. Med. Assn. 81:651.

————: 1933. The Massachusetts plan for the eradication and control of infectious laryngotracheitis. Jour. Am. Vet. Med. Assn. 83:214.

————: 1934. Infectious laryngotracheitis vaccination. Mass. Agr. Exper. Sta., Bul. 311.

Hinshaw, W. R.: 1931. A survey of infectious laryngotracheitis of fowls. Calif. Agr. Exper. Sta., Bul. 520.

————, Jones, E. E., and Graybill, H. W.: 1931. A study of mortality and egg production in flocks affected with infectious laryngotracheitis. Poultry Sci. 10:375.

Hudson, C. B., and Beaudette, F. R.: 1932a. Infection of the cloaca with the virus of infectious bronchitis. Science 76:34.

———— and Beaudette, F. R.: 1932b. The susceptibility of pheasants and a pheasant-bantam cross to the virus of infectious bronchitis. Cornell Vet. 22:70.

Kernohan, G.: 1930. Infectious bronchitis in fowls. Calif. Agr. Exper. Sta., Bul. 494.

Komarov, A., and Beaudette, F. R.: 1932. Carriers of infectious bronchitis. Poultry Sci. 11:335.

May, H. G., and Tittsler, R. P.: 1925. Tracheo-laryngitis in poultry. Jour. Am. Vet. Med. Assn. 67:229.

Schalm, O. W., and Beach, J. R.: 1935. The resistance of the virus of infectious laryngotracheitis to certain physical and chemical factors. Jour. Infect. Dis 56:210.

Seddon, H. R., and Hart, L.: 1935. The occurrence of infectious laryngotracheitis in New South Wales. Australian Vet. Jour. 11:212.

————: 1936. Infectivity experiments with the virus of laryngo-tracheitis of fowls. Australian Vet. Jour. 12:13.

Seifried, O.: 1931. Histopathology of infectious laryngo-tracheitis in chickens. Jour. Exper. Med. 51:817.

# CHAPTER TWENTY-ONE

# AVIAN PNEUMOENCEPHALITIS (NEWCASTLE DISEASE)

By J. R. BEACH, *Department of Veterinary Science,*
*University of California, Berkeley, California*

☆ ☆ ☆

Doyle, in 1926, encountered in England what he termed "a hitherto unrecorded disease of fowls due to a filter-passing virus," for which he proposed the name Newcastle disease because the first outbreaks occurred at that city on the river Tyne (Doyle, 1927). During the ensuing years a disease of poultry which had previously been reported from the Dutch East Indies and which was encountered in various parts of the Orient, on certain South Pacific islands, in Australia, Palestine, Kenya Colony, and more recently in Italy, Germany, and French Equatorial and South Africa was proved to be identical with Newcastle disease. The disease in the various locations where it was encountered was designated by a variety of names which included pseudo-fowl pest, Philippine fowl disease, Chosen disease, Madras fowl pest, and Ranikhet disease. Doyle (1933) proposed that, to avoid confusion from such a plurality of nomenclature, the disease be universally termed Newcastle disease, although admitting that this designation was obviously unsuitable for a disease with such widespread distribution. It is to be noted that the disease had not been identified in any part of the Western Hemisphere.

In the reports concerning Newcastle disease mentioned above, it is described as being readily transmissible by inoculation of healthy chickens with tissues of diseased ones and as causing severe losses from death. Doyle (1927), for example, states that the mortality in naturally infected flocks is usually 100 per cent; and Dobson (1939) describes an outbreak in which "the disease spread rapidly and, in a few days, 5,000 had died or had been destroyed as affected . . . . "; Stater (1945) reports that it is the most dreaded disease of chickens in India; and Beaudette (1946) refers to "the nearly 100 per cent mortality resulting from the infection." Further description of the disease as it has occurred outside of the United States is not given in this chapter. Readers who are interested are referred to the comprehensive review of the literature on Newcastle disease by Beaudette (1943).

The term, avian pneumoencephalitis, first appeared in the literature in 1942 in reports by Beach (1942b) on a disease of poultry which had been

prevalent in California for several years. The events leading up to this and which subsequently linked this disease with Newcastle disease are briefly as follows:

In 1940, numerous flocks in California from two to ten weeks old were affected with a type of disease which had not been observed previously (Beach, 1940). The outbreaks began as a respiratory trouble which spread rapidly through the flocks. In a few days after its appearance, some of the chicks developed symptoms of involvement of the central nervous system, the number of such cases varying from less than 1 per cent to about 50 per cent in different flocks. The disease became known as a respiratory-nervous disorder (Stover, 1942a; Beach, 1942a) but this was soon replaced by the term avian pneumoencephalitis (Beach, 1942b). Despite the fact that the spread of the disease through a flock was very rapid, artificial transmission proved difficult, and it was not accomplished until 1941. The cause of the disease was then shown by Stover (1942b) and Beach (1942a, 1942b) to be a filtrable virus which could be propagated in chicken embryos. The virus in 1942 was also identified with a previously unclassified type of respiratory disease of laying pullets, which had been prevalent in the state since 1935 or earlier and which was known in different localities as "chicken-flu" and "nine-day pneumonia." The loss from death in infected flocks had been so small that poultry raisers felt concern about the disease only because of its depressant effect on egg yield. In 1942, however, outbreaks in which some of the birds showed nervous symptoms were observed (Beach, 1942b; Stover, 1942c). The disease has continued to be prevalent throughout the state in growing chicks and in pullets preceding and after they have reached laying age. Some outbreaks have been accompanied by a disturbingly high mortality but the major loss caused by it has been from temporarily retarded development of the young and reduced egg yield of flocks which were laying when they became affected.

In culturing pneumoencephalitis virus in chicken embryos it was found that, irrespective of the severity of the disease in the naturally infected chicken from whose tissues the virus had been isolated, its virulence so increased that inoculation of young chickens with minute amounts of infected embryo tissues or fluids would produce an acute fatal infection and types of lesions, particularly hemorrhages of various portions of the digestive tract, which are seldom if ever seen in naturally infected birds. These findings suggested a possible relationship between the virus and the viruses of either Newcastle disease or fowl plague, neither of which was known to exist in the United States. To explore this possibility, Newcastle disease and fowl plague antiserums were obtained from England in 1943, through the cooperation of the Bureau of Animal Industry, United States Department of Agriculture, and used in neutralization tests with cultured pneumoen-

cephalitis virus. The infectiveness of the virus was not affected by the fowl plague antiserum but definite neutralization of the virus was obtained with the Newcastle disease immune serum (Beach, 1944a). These results indicated that the virus of pneumoencephalitis is immunologically identical with the virus of Newcastle disease. This finding was later confirmed by Brandly and co-workers (1946a) by means of both neutralization and cross-protection tests. The interpretation of this finding has been a somewhat controversial matter. The question involved was well expressed in the report of the committee on transmissible diseases of poultry (Stafseth *et al.*, 1944) at the forty-eighth annual meeting of the United States Livestock Sanitary Association which reads in part: "An important development in the field of poultry pathology . . . . is a report by Beach on the neutralization in vitro of avian pneumoencephalitis virus by Newcastle disease immune serum. . . . . whether this means that Newcastle disease and pneumoencephalitis are caused by the same virus or by two antigenically related viruses remains to be proved." Factors which would seem to support the latter conception are the differences between the clinical aspects of Newcastle disease and the ease with which it can be transmitted from field cases by artificial means, as given in reports concerning it in other countries, and pneumoencephalitis in the United States. Newcastle disease is described as causing nearly 100 per cent mortality and as being readily transmissible by inoculation with tissues of naturally infected birds. In contrast, transmission of pneumoencephalitis to normal chickens has proved difficult, and the average mortality from it has been small. Furthermore, during the more than ten years of its known existence in California it has shown no sustained tendency to assume a more severe character. In the absence of more conclusive evidence to the contrary, however, equal credibility must be assigned to the concept that the disease in the United States is Newcastle disease which has for some undetermined reason occurred in a benign form up to the present time, but which at any time may take on the devastating characteristics which Newcastle disease has exhibited in other parts of the world. Only time will reveal which of the opposing concepts is correct. In the meantime, the writer prefers to use the term pneumoencephalitis for the disease in the United States because, if for no other reason, this term should make it clear that it refers to the type of disease which thus far has been experienced here.

The first indication of the presence of pneumoencephalitis infection in any state other than California was the report by Minard and Jungherr (1944) that they had demonstrated significant concentrations of neutralizing antibodies for a California strain of pneumoencephalitis virus in the serum of "normal" chickens in Connecticut. The donors of the serum were from the University of Connecticut flocks in which, presumably, pneumoencephalitis infection had never been present. According to the tenets of

virology, however, the birds with a high level of pneumoencephalitis neutralizing antibodies must have had past contact with the virus, but the manner in which this could have occurred was not determined. From subsequent experience with the disease, however, it is known that the flock could have been affected with a subclinical type of disease without its presence being suspected.

In November, 1944, the disease appeared among pullets at the Western Washington Agricultural Experiment Station. The diagnosis was made by serological tests and challenge with virus of birds shipped to the Department of Veterinary Science, University of California. In a later description of the outbreak (Berg, Bearse, and Hamilton, 1947) no mention is made of a probable source of the infection or of any loss having resulted except from decreased egg yield and an adverse effect of the infection on egg quality.

During the winter of 1944–45 the disease was recognized in New Jersey (Beaudette and Black, 1945) in both laying flocks and chicks. As had happened in California several years earlier, because of the low mortality and the respiratory symptoms being mistaken for infectious bronchitis, the disease was well established in the state before it was identified as pneumoencephalitis. Recognition of the disease in neighboring states quickly followed.

It was now realized that pneumoencephalitis might be more widely distributed than had been believed and might become a nationwide poultry disease problem. Consequently, under the leadership of the United States Bureau of Animal Industry, conferences of research workers, livestock disease control officials, and representatives of the poultry industry were held to devise ways and means of attacking the problem. An important outcome of these conferences was an arrangement whereby material from suspected cases in any state could be submitted for diagnosis to certain designated laboratories having adequate diagnostic facilities. Full advantage has been taken of this with the result that up to the present (November, 1947) the disease has been identified in forty-three states, the District of Columbia, and the Territory of Hawaii. In some instances there was good evidence of the infection having been introduced from another state by live birds. In many cases, however, the source of the infection could not with certainty be traced, and definite determination as to whether or not the outbreak detected represented the first occurrence of the disease in the state could not be made.

**Etiology.** The cause of pneumoencephalitis is a filtrable virus which immunologically is identical with the virus of Newcastle disease. The virus has been shown to be present in the air sac membranes, tracheal exudate, brain, spleen, and lungs during the early stages of the disease, especially in severely infected chickens. Much difficulty may be experienced in demon-

strating the virus in tissues of cases of a few days' standing. It grows readily in chick embryos, a characteristic which has greatly aided in the detection of the disease. Virus in lung tissue dried while frozen or placed in 50 per cent glycerine and stored in the refrigerator has remained viable for 195 days and 85 days, respectively, the longest periods tested (Beach, 1942b). Allantoic fluid of infected embryos stored in a dry ice refrigerator remained infective for as long as two years (Beach, unpublished data). Jungherr (1948) found that cultured virus on burlap strips retained virulence for 13, 25, and 55 days at temperatures of 29° F., 51° F., and 72° F., respectively. Evidence that the virus survives for only a brief period in an environment contaminated by naturally infected chickens, however, is provided in the report by Jungherr on failure of susceptible chickens to become infected when placed in uncleaned or cleaned but non-disinfected houses immediately after infected chickens had been removed from them (Jungherr, 1948).

**Symptoms in chicks.** The majority of outbreaks of pneumoencephalitis in chicks begins as a respiratory trouble, indistinguishable from infectious bronchitis; but in 1 or 2 days, or in some instances after a longer interval, some of the chicks may develop symptoms of involvement of the central nervous system. In other outbreaks, however, respiratory symptoms alone are seen. The respiratory phase has usually terminated within one to three weeks. The nervous symptoms, however, have been seen over a longer period.

The respiratory symptoms are gasping, coughing, and râles, and many of the affected chicks emit a peculiar rapid low chirp.

The symptoms from involvement of the nervous system are quite varied in different individuals. (See Fig. 21.1 A, B, C, and D.) They consist of motor ataxia, partial or complete paralysis of one or both legs, tremor of the head or whole body, and incoordination of action of the neck muscles, as a result of which the head may be drawn straight back between the shoulders, downward and backward toward the breast, twisted to either side, or drawn to the right or left. The chickens may walk in circles or backwards. The appetite of birds thus affected may be unimpaired, and notwithstanding the inability to control muscular movement many chickens will manage to consume enough food to keep, themselves fairly well nourished. On the other hand, affected chickens may become droopy, eat little or no food, and quickly become emaciated and weak. In cases of fatal infection, the chickens are likely to become prostrated, show clonic spasms or a rhythmic twitching of parts or all of the body, and go into a state of coma before death. In numerous experimentally produced cases, death has occurred in less than 24 hours after the first symptom was observed. Although some chickens have recovered from the nervous affliction, the majority of those which escaped death have retained the symptoms indefinitely.

The most common findings on post-mortem examination of chicks are cloudiness or thickening of the air sacs and, in many cases, also of the thin membranes of the abdominal cavity with a film of yellowish exudate, and varying amounts of yellowish or clear mucus in the trachea and large bronchi. Many infected chicks, however, particularly those with nervous symptoms alone, have shown no visible lesions. Submucous petechial or more diffuse hemorrhages of various portions of the intestinal tract, the proventriculus in particular, are common post-mortem findings in acute cases produced by inoculation with cultured virus, but such lesions have very rarely been seen in infected chicks on farms.
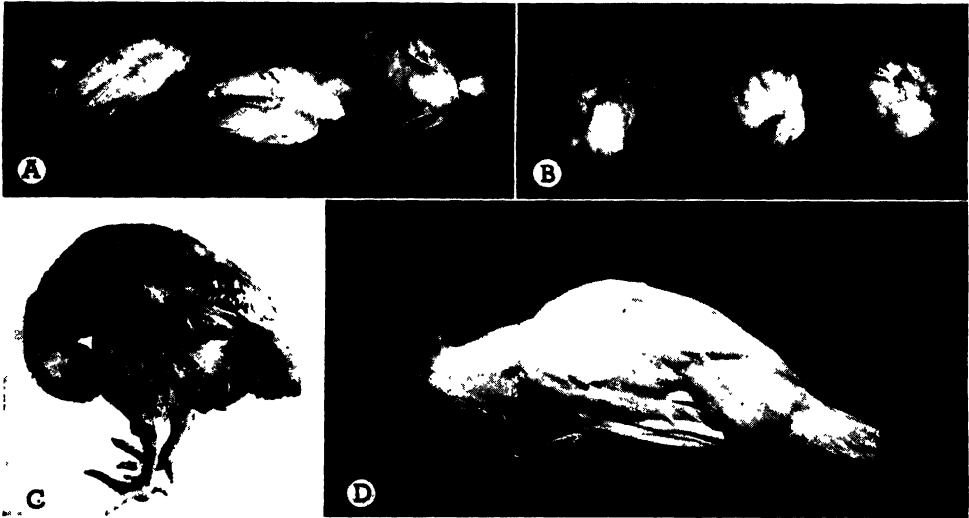


Fig. 21.1. A—Pneumoencephalitis. Natural infection. Right and left paralysis of legs. Center, paralysis of legs and wings. B—Pneumoencephalitis. Natural infection. Paralysis of legs and incoordination of neck muscles. C—Pneumoencephalitis. Natural infection. Incoordination of neck muscles. (Photo courtesy of H. A. Hoffman.) D—Pneumoencephalitis. Artificial infection. Incoordination of neck muscles. (Photo courtesy of D. E. Stover.)

As was stated before, both respiratory and nervous symptoms have been present in nearly all outbreaks of the disease among chicks. The respiratory phase has often affected nearly 100 per cent of a flock, the nervous phase from 1 to 45 per cent. In a few instances, however, respiratory symptoms either have been absent or so very mild that they escaped detection. The nervous involvement tends to be more rapidly fatal in the younger chicks. The average mortality is probably between 5 and 10 per cent. A greater loss, however, may result from unthriftiness of the survivors. Most of the flocks affected have been from three to ten weeks old, although outbreaks among younger chicks have been observed. The disease has appeared only once on some farms; on others it has recurred in successive broods of chicks.

In pullets from three to five or five and one half months old; i.e., up to the start of egg production, the disease tends to be less severe than in younger birds, especially with respect to the number showing nervous symptoms. Respiratory symptoms may be quite pronounced, but some outbreaks have been observed in which so few birds were coughing or gasping that they were difficult to detect and the principal evidence of illness was slightly decreased appetite. There have been instances, however, in which marked depression and inappetence were the predominant symptoms and numerous birds developed nervous symptoms which caused either their death or removal as culls.

**Symptoms in laying flocks.** Pneumoencephalitis in laying flocks is characterized by a sudden onset and rapid spread. The earliest symptoms in most outbreaks are coughing and gasping of a few or practically all of the birds. In some instances, however, respiratory symptoms are either absent or so obscure that they escape detection. These symptoms are indistinguishable from those of infectious bronchitis and may be confused easily with those of laryngotracheitis. Food consumption decreases rapidly, in some instances to the point that within a week or less the birds will eat no mash at all. Accompanying the decline in appetite is a drop in egg production so rapid that within 5 to 10 days, in some instances, no eggs are being laid. The respiratory symptoms may have disappeared by this time. The appetite rapidly returns to normal, egg yield improves and may attain the pre-infection rate within two to four weeks. During this period of about 30 days, the outbreak may have run its course in a pen of birds without any mortality attributable to the disease having occurred, the only loss being from the depressed egg yield. The infection, as a rule, quickly appears in other pens and houses until all susceptible groups become affected.

Symptoms, in addition to those described above, which are seen in more severe outbreaks are as follows: Depression or droopiness of a few of the birds is seen at the onset of an outbreak. The number showing these symptoms increases rapidly, and within a week practically the entire flock may be sitting around on the roosts or floor and eating little or nothing at all. Usually a number of floor or yard eggs, many with soft or imperfectly formed shells, are laid. Recovery may be so rapid that marked improvement in the condition of the flock may be apparent in 24 to 48 hours. A variable number of the birds may develop nervous symptoms consisting of torticollis and other types of muscular incoordination, paralysis of one or both legs or wings, and clonic spasms which may involve only a leg, wing, or the neck, combinations of these parts, or the whole body. The clonic spasms may perhaps be considered as particularly characteristic of pneumoencephalitis. Birds thus affected may not die but may as well be considered as "mortality" because the damage to the nervous system is likely to be permanent. The

number of such cases determines to a large degree the amount of mortality that occurs. This varies between wide limits both in different flocks and in different pens of birds of the same age on one farm. To cite extreme examples: Practically all of an infected flock of 750 laying pullets showed marked depression, but only one of them developed nervous symptoms, and this was the only one lost by death. The total mortality of an infected flock of 1,650 birds divided into five pens was 15 per cent. The variation of the mortality in the different pens ranged from a low of 2 per cent to a high of 82 per cent. The improvement of a severely affected flock with respect to appetite and general appearance may be spectacularly rapid. Egg production, however, increases more slowly and 30 to 60 or more days may elapse after the onset of the disease before the birds are laying at the pre-infection rate. If a molt is induced by the effects of this disease, as may happen if flocks of laying pullets become infected in late fall, the return of normal egg yield will be even further delayed.

Autopsy reveals no lesions that are pathognomonic of pneumoencephalitis. Some fluid, mucus, or caseous exudate may be present in the trachea, and the air sac membranes may be clouded and contain some yellowish exudate. Liquid or caseated egg yolk may be found in the abdominal cavity. In many cases, however, no gross lesions can be detected. Autopsy findings, therefore, are of little diagnostic aid.

**Subclinical infection.** An insidious characteristic of pneumoencephalitis is that its visible effect on individuals or a flock may be so slight that its presence is not suspected. Infection of this type, which has also been termed inapparent or asymptomatic, is detected by means of serological tests (hemagglutination-inhibition and serum neutralization) and by inoculating the birds with virulent virus. It has been detected in many individuals being used experimentally and as a generalized infection of farm flocks. The latter was demonstrated in closely observed laying flocks on a group of neighboring farms (Beach, 1944, unpublished data) on which the disease had occurred in 1942, was apparently absent in 1943, and recurred in an unusually severe form in the fall of 1944, the disease being confined to pullets hatched in the spring of that year. The hens, however, which had been on the farms as pullets in 1943 and among which no evidence of pneumoencephalitis had ever been observed were not affected. Representative specimens of these flocks gave positive reactions to the hemagglutination-inhibition and serum neutralization tests with pneumoencephalitis virus and were refractory to challenge. These results indicate that the hens had become immunized by past subclinical infection.

Another interesting example of subclinical infection was detected in a large breeding flock which had exceptionally close supervision (Bankowski, 1946, unpublished data). No disease condition suggestive of pneumoen-

cephalitis had ever been observed in this flock. In January, 1946, the occurrence of several cases of mild conjunctivitis and lachrymation led to specimens being submitted to a diagnostic laboratory. These came at a time when all specimens received were routinely being given hemagglutination-inhibition tests for pneumoencephalitis for the purpose of determining the reliability of the test as a diagnostic procedure. The serums of these birds gave a strong positive reaction to the tests, and the birds which were challenged proved refractory to pneumoencephalitis virus. Tests of additional birds from other groups on the farm revealed that subclinical pneumoencephalitis infection had been present throughout the breeding establishment. It is of interest that clinical pneumoencephalitis did not occur on the farm until the fall of 1947, and then it was restricted to the first- and second-year progeny of the breeding birds among which the subclinical disease had been detected early in 1946. Jungherr and Terrell (1946) also report having detected several cases of subclinical infection and of the spread of the disease in subclinical form on a farm for two or three months following an active outbreak. Pneumoencephalitis occurring without its presence being suspected adds to the problem of determining how widely the disease is distributed. It also would be a handicap to the success of a program of control and eradication of the disease by livestock disease control officials.

**The disease in turkeys.** Numerous flocks of turkeys have been affected, but for reasons as yet unknown outbreaks have been less prevalent in birds of this species than among chicken flocks in the same area. The symptoms in turkeys do not differ essentially from those of affected chickens. The respiratory symptom which seems to be rather typical for turkeys is an intermittent hoarse cough or sneeze. Gasping and other indications of respiratory involvement as described for chickens, however, have also been observed in turkeys. The number of cases showing nervous symptoms may vary from none to a relatively large proportion of the flock. In one outbreak which occurred in September in a flock being raised for the Thanksgiving market, 25 per cent of the birds developed nervous symptoms. Very few of these were fatally affected, but all were lost to the grower because they were unacceptable to buyers. Fenstermacher and co-workers (1946) state, however: "Nearly mature turkeys seldom show nervous symptoms. As a rule, the owner is unaware that anything is wrong."

**Transmission.** The exact sources of the infection which were responsible for initial outbreaks of the disease in previously noninfected flocks or areas are for the most part rather obscure. Recent observations and experimental studies, however, have yielded considerable information on this point.

In a number of instances live birds from previously infected flocks have been rather definitely incriminated as having carried the infection to other flocks to which they were added for breeding purposes. The possibility of

recovered birds being healthy virus-carriers was demonstrated by Beach (1942b) by the isolation of the virus from the lungs of chickens two to three months after recovery from the disease. However, Beach (unpublished data) has failed in numerous attempts to transmit the disease from naturally infected birds, showing nervous symptoms, to normal chickens by contact exposure in cages. A similar transmission trial by Levine (cited by Jungherr, 1948) likewise resulted in failure. Possible transmission of the disease through hatching eggs from infected flocks is suggested by the isolation of the virus from ovarian tissues (Beach, 1942, unpublished data), from fresh eggs of infected hens (Van Roekel, 1946; Jungherr, 1946), and by the occurrence of the disease in very young chicks. Further supporting evidence is provided in the report by DeLay (1947) of having isolated the virus from the yolk sac of 4-day-old chicks, dead embryos, and infertile eggs from parent stock known to be infected with pneumoencephalitis. The chicks were hatched from eggs obtained when egg production was markedly depressed. The embryos from the same parent stock were from eggs collected during the recovery period. The infertile eggs were from a different infected flock. In this case the virus was not demonstrated in living or dead embryos or from the chicks which hatched. While these findings strongly support the possibility that hatching eggs may be a medium of transmission of the disease from breeding hens to their offspring, evidence is lacking that this could occur after a flock has fully recovered. In fact Brandly, Moses, and Jungherr (1946) demonstrated the presence of antiviral antibodies rather than virus in the yolk of eggs laid by immune recovered hens. During incubation of such eggs, the developing embryos gradually acquired the virus-neutralizing property from the yolk. The blood serum of the chicks on hatching had virus-neutralizing ability, and the chicks were refractory to inoculation with virus. There was a rapid decline in both neutralizing titer of the serum and resistance of chicks to the virus after the second week, only a small proportion remaining refractory to infection for five or six weeks. The inoculation with virus did not produce lasting active immunity in the passively immune chicks. Under farm conditions, such chicks would be refractory if exposed to the disease during the first month after hatching, but could become infected if exposed again when they were older. This passive immunity would also interfere with development of immunity from vaccine administered to chicks under a month old.

Cultured pneumoencephalitis virus has been transmitted to susceptible chickens by exposing them to artificially contaminated air (DeOme, 1947, unpublished data). From this, together with field observation, it has been assumed that the disease may be transmitted by the air-borne route under natural conditions, although the actual presence of the virus in such air had not been demonstrated. Recently, however, DeLay, DeOme, and Bankowski (1947) obtained proof of the infectivity of the air of poultry

houses containing birds with natural pneumoencephalitis infection. These investigators obtained samples of air at the level of the heads of the chickens in three poultry houses each containing an infected flock, either by drawing it through allantoic fluid of normal embryos or collecting the floating dust with a vacuum cleaner. Pneumoencephalitis virus which was infective for either chickens or embryos was demonstrated in the material thus collected after it had been rendered bacteriologically sterile by treatment with penicillin and streptomycin. A direct test of the infectivity of the air for chickens was made by confining susceptible chickens in cages suspended 4½ feet above the floor where they were out of contact with any contaminated material which was not air-borne. All developed respiratory symptoms within 6 days; their serum was shown to contain hemagglutinating-inhibition antibodies in significantly high concentration on the eighth day, and on the fifteenth day the birds proved refractory to a challenge dose of cultured pneumoencephalitis virus. Such demonstration of the virus in the air of poultry houses makes it appear possible that air-borne transmission is largely responsible for the rapid spread of the disease through a poultry house, from house to house on a farm, and with dust carried by the wind or other means from farm to farm in congested poultry districts.

**Effect on egg quality.** Lorenz and Newlon (1944) reported studies of this phase of the pneumoencephalitis problem after having received field reports of abnormalities in fresh-laid eggs following outbreaks of the disease. Their findings were, for the most part, obtained from 100 trap-nest pullets which they had under observation immediately preceding and for 45 days following the onset of the disease. The outbreak was characterized by respiratory symptoms, profound depression, and marked reduction in egg yield. The mortality, however, was nil. The abnormalities found were eggs containing no true air cell but instead an accumulation of small free-floating air bubbles and poor albumen and shell quality. These abnormalities were observed throughout the 45-day period but progressively decreased toward the end. A total of 1,294 eggs were laid. Of these, 6.4 per cent had bubbly air cells, all of which were laid by 21 per cent of the birds. The incidence of bubbly air cells bore no direct relation to the severity of the disease. Approximately the same number of eggs had abnormal shells and these were laid by 44 per cent of the flock. Decreased albumen quality was detected in nearly all eggs produced. Pneumoencephalitis was said to be the first factor, other than the age of the bird, that has been shown to modify the albumen height of fresh-laid eggs. Similar findings were reported by Berg, Bearse, and Hamilton (1947) from studies made subsequently.

**Animals susceptible.** Beach (1942b; 1943, unpublished data) found Chinese and silver pheasants, California quail, and chukar partridges as susceptible as young chickens to inoculation with cultured pneumoencephalitis virus. Pigeons were susceptible only to larger doses than are re-

quired to infect chickens. Levine *et al.* (1947) reported a severe outbreak of the disease among young artificially hatched and reared Chinese pheasant chicks. Fenstermacher *et al.* (1946) added Hungarian partridges and ring doves to the list of susceptible birds but failed to confirm the findings of Beach and Levine with respect to the high susceptibility of Chinese pheasants and chukar partridges. Beach and Bankowski (1947, unpublished data) found that wild Japanese doves (*Geopelia striata*) were highly susceptible to the cultured virus. These birds were trapped around the houses of a poultry farm on which an outbreak of pneumoencephalitis had recently occurred.

Beach (1942b) and Brandly and co-workers (1946) reported that mice were refractory to the virus, and the former reported that attempts to infect guinea pigs did not give clear-cut results. The successful infection of Syrian hamsters by intracerebral inoculation with a California strain of virus maintained in chicken embryos has been reported by Reagan, Lillie, Poelma, and Brueckner (1947a, 1947b). The disease has been carried in these animals through a series of fifty passages. The symptoms shown by affected animals are those of a central nervous system disturbance. Virus has been demonstrated only in the brain tissue. Attempts to establish the hamster-adapted virus in mice, guinea pigs, and rabbits are reported as unsuccessful. These investigators are hopeful that the hamster-adapted virus will become so modified that it can be utilized for vaccinating chickens. Reagan *et al.* (1947b) report having produced nervous symptoms in a rhesus monkey by intracerebral injection of the hamster-adapted virus but failed to do so with embryo propagated virus. DeLay (1946, unpublished data), however, reports having produced fatal infection in two monkeys by intracerebral injection of cultured virus and that the brain tissue of one of them was infective for chicks. In neither instance, however, were subsequent passages made to determine whether the infection could be maintained by serial passages in this species of mammal.

**Diagnosis.** It is recognized generally that pneumoencephalitis usually cannot be definitely recognized by means of the clinical picture and/or the post-mortem findings. The respiratory symptoms cannot be distinguished from those of infectious bronchitis or from laryngotracheitis if the blood which is usually mixed with the tracheal exudate seen with the latter disease is absent. The cloudiness of the air sac membranes which may be found on autopsy is suggestive of pneumoencephalitis, but this condition is neither pathognomonic nor of regular occurrence. A respiratory disease in a laying flock accompanied by decrease in egg production may be either pneumoencephalitis or infectious bronchitis but is more likely to be the former if the egg production drops rapidly and nearly or entirely ceases within a few days. However, respiratory symptoms associated with or followed by nervous symptoms in the case of young birds or, in a laying flock, respiratory

symptoms accompanied by a sudden sharp decline in egg yield, depression, loss of appetite especially for mash, and nervous symptoms in some birds, can with considerable assurance be classified as pneumoencephalitis. If there is any question whatsoever as to a positive diagnosis, the proper procedure is to submit live specimens, or tissues or blood serum from them to a laboratory having adequate facilities for identifying the disease. The laboratory procedures which may be employed are (1) isolation of the virus by inoculating chicken embryos with tissues or tracheal exudate, (2) conducting hemagglutination tests with blood serum, (3) conducting neutralization tests with blood serum, and (4) testing live birds for immunity by inoculating them with a challenge dose of cultured virus. For the first of these, birds in the early stages of the disease and preferably not showing nervous symptoms should be selected; for the other three, birds which have been ill for at least a week are required; specimens showing nervous symptoms but which otherwise appear to be in a fair state of health are particularly satisfactory.

**Virus isolation.** Specimens for virus isolation attempts should, as stated above, be living and in the early stages of disease. If location permits, the birds should be taken (not shipped) directly to the diagnostic laboratory. Otherwise tissues should be removed aseptically, placed in sterilized containers and shipped to the laboratory. Suitable tissues are brain, spleen, and lung and also yolk if a laying bird is selected. Tissues should either be frozen and delivered to the laboratory in that state or preserved in a 50 per cent glycerine solution, the dilution if possible being made with phosphate buffer adjusted to pH 7.4. The glycerine-preserved tissues should be refrigerated until shipped, and packed so that the temperature will not rise appreciably during transit. Three or more 8- to 10-day-old embryos are inoculated with sterile suspensions of one or all of the tissues. If the virus is present, some or all of the embryos should die within 6 days. Embryos killed by the virus regularly show marked congestion. The hemagglutination test, which is described later, is a valuable aid in determining whether death of embryos is due to virus infection. It is often desirable, however, to employ the inoculation of susceptible young chickens with tissue and/or fluids of dead embryos to obtain positive proof of the presence of the virus. In many instances a diagnosis can be made from the results of the initial embryo passage, but frequently additional passages are required. The direct inoculation of chickens with tissues of field cases, as a diagnostic procedure, is so uncertain of success that it is seldom attempted. J. P. Delaplane has developed an embryo-inoculation technique which he claims is being routinely and successfully used for the differential diagnosis of infectious bronchitis and pneumoencephalitis. The inoculum is a saline suspension of exudate removed from the trachea of birds having recently acquired infection. Contaminating bacteria are destroyed, without any virus present being affected, by the addition of 0.025 grams of streptomycin to each cc. of exudate sus-

pension. The mixture is well shaken and used immediately for inoculating into the allantoic cavity of the embryos. The presence of infectious bronchitis virus is indicated by dwarfing, but not death, of the embryos. This dwarfing effect is said to be detectable at the first embryo passage and to become more distinct with one or two additional passages. If the inoculum contains pneumoencephalitis virus, one or more of the embryos, as stated above, will die within 6 days and usually within 48 to 72 hours.

**Hemagglutination and hemagglutination-inhibition tests.** The phenomena of the agglutination of chicken red blood cells by influenza virus in allantoic fluid of infected embryos and the inhibition of the red cell agglutination by serum of immune animals were first described by Hirst (1941, 1942). Later Burnet (1942) and Lush (1943) reported that red cells were agglutinated respectively by an Australian strain of Newcastle disease virus and European strains of both Newcastle disease and fowl plague. Hemagglutination and hemagglutination-inhibition by pneumoencephalitis virus and immune serum, respectively, were demonstrated by Beach (1947) and Brandly *et al.* (1946a) in 1944. Since that time both the HA (hemagglutination) and the HI (hemagglutination-inhibition) tests have been extensively employed as diagnostic aids for pneumoencephalitis, the former for determining the presence and identification of the virus in allantoic fluid of infected embryos and the latter for testing the serum of infected or recovered birds for the presence of hemagglutination-inhibiting antibodies.

The techniques of the tests vary somewhat as to detail but not fundamentally in different laboratories. Techniques currently employed at the Department of Veterinary Science, University of California, are, briefly, as follows: In the HA test, 0.5 cc. amounts of two-fold saline dilutions of virus; i.e., the allantoic fluid of infected embryos, ranging from 1:10 to 1:1,280 or higher are made in a series of chemically clean agglutination tubes. The tubes are placed in racks with coarse mesh wire or glass bottoms so that the bottoms of the tubes are clearly visible. Five-tenths cc. of a 0.75 per cent suspension of washed chicken red cells is added to each tube. A control tube containing 0.5 cc. each of saline and red cell suspension is added and the rack well shaken. After 25 to 35· minutes at room temperature, readings are made by viewing the pattern formed by the sedimented cells on the bottom of the tubes. Readings are facilitated by placing the rack beneath a fluorescent lamp and above a mirror set at a 30-degree angle, and viewing the images on the bottom of the tubes in the mirror (Fig. 21.2). When the cells are not agglutinated, the sedimenting cells form small discs of gradually increasing size at the lowest point of the curvature of the bottom of the tubes. When agglutination occurs, the sedimenting cells form either a thin uniform blanket covering the entire bottom of the tube ($+ + + +$ reaction); a thicker blanket with an irregular outline covering a portion of the bottom of

the tube (+++ reaction) ; a small central disc surrounded with a granular area of agglutinated cells (++ reaction) ; or a larger central disc surrounded with a narrow granular ring of agglutinated cells (+ reaction). The appearance of sedimented cell patterns on the bottom of the tubes is shown in Figure 21.3. The highest dilution of virus which gives at least a +++ reaction is termed the hemagglutination titer of the virus, and the amount of virus which produced this reaction is termed a unit. The agglutinated
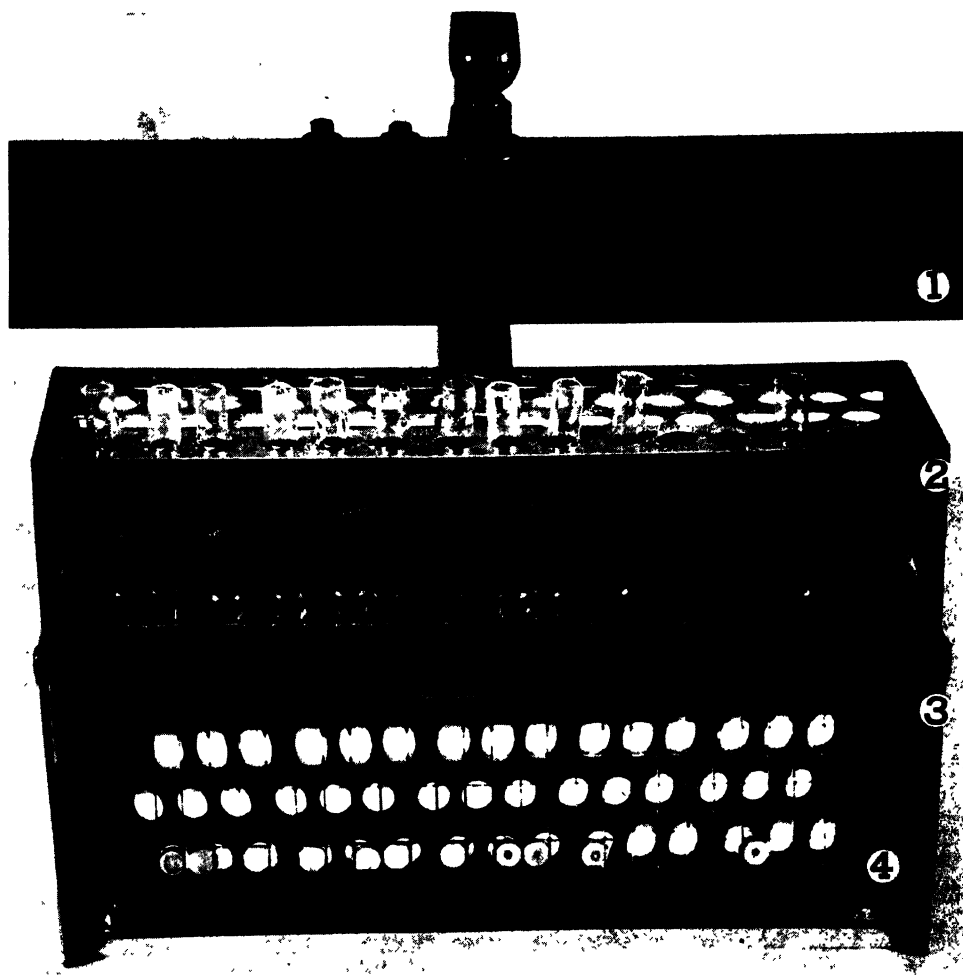


FIG. 21.2. Apparatus for making the hemagglutination and hemagglutination-inhibition tests and for reading the reactions. (1) Adjustable two-tube flourescent lamp. (2) Metal rack accommodating four rows of fifteen tubes. (3) Metal support for rack with three sides closed to keep out light. (4) Mirror set at a 30° angle for viewing appearance of bottoms of the tubes. The clear oval mirror images are those of the unfilled holes in the rack. Tubes 1–10 in the bottom row are described in Figure 2. Tube 11 is a control containing only red blood cells and saline. Photo taken 30 minutes after the test was set up.

DILUTIONS OF VIRUS

1/5  1/10  1/20  1/40  1/80  1/160  1/320  1/640  1/1280  1/2560
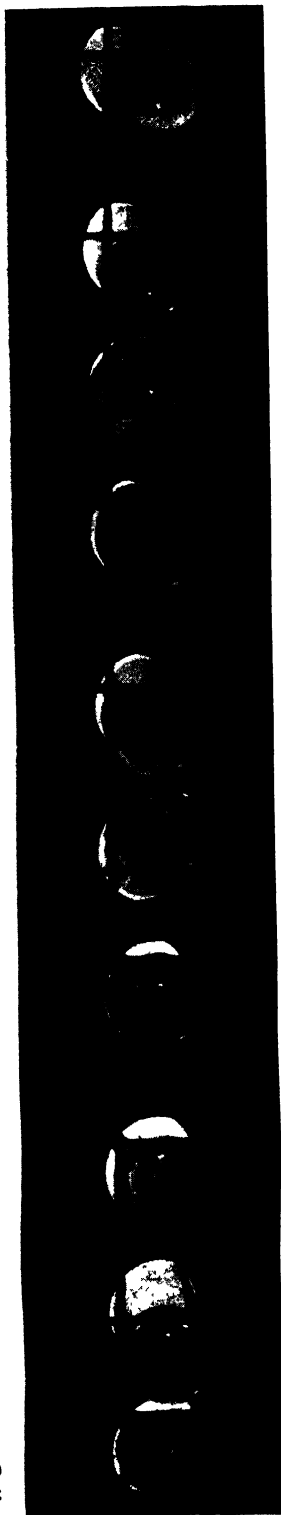
1  2  3  4  5  6  7  8  9  10



FIG. 21.3. Titration of virus by the hemagglutination test. Mirror images of the bottoms of the tubes. Red cells agglutinated in tubes 1–7 and have settled to form a thin blanket on the bottom of the tubes. The hemagglutination titer of the virus is expressed as 1:320. In tube 1 the agglutinated cells have begun to slide toward the center. No agglutination in tubes 8–10. The cells in settling have formed a disc at the lowest point. The clear spot in the center of the disc of cells in tube 10 is due to irregularities in the curvature of the bottom. Photo taken 30 minutes after test was set up.

cells soon begin to slide toward the lowest point of the tube and, in a short time, form a disc which cannot be distinguished from a negative reaction.

In the hemagglutination-inhibition test, a preliminary step is the determination of the hemagglutination titer of the virus. This must be done each day and with each lot of virus used. Next, 0.25 cc. amounts of twofold dilutions of serum from a suspected pneumoencephalitis case, ranging from 1:10 to 1:1,280 or higher are made in a series of tubes. The next step is the addition to each tube of diluted serum 0.25 cc. of diluted virus. The dilution used is two twofold dilutions below the highest dilution which had given a +++ or ++++ reaction when the virus was titrated. For example, if the HA titer of the virus is 1:320, a 1:80 dilution is used in the HI test. The 0.25 cc. amount of this dilution is considered to contain two units of virus. After thorough shaking, the mixtures of serum and virus are allowed to stand for 10 minutes after which 0.5 cc. of the red cell suspension is added to each. A normal-serum control series of tubes and a tube containing 0.25 cc. each of serum and saline and 0.5 cc. of red cells, and one containing 0.5 cc. each of saline and red cell suspension are included. Readings are made in 25 to 35 minutes. If the serum contains no HI antibodies, a ++++ or +++ plus HA reaction occurs in all tubes of the series. This is a *negative* HI reaction. On the other hand, when the serum contains HI antibodies, and agglutination of the red cells is inhibited in one or more of the tubes of a series, the next one or two tubes may show a ++ or + HA reaction, and the remainder of the tubes a +++ or ++++ HA reaction. This is termed a positive HI reaction. The highest dilution of serum which inhibits hemagglutination is termed the HI titer of the serum. An alternative procedure for setting up HI tests employed in many laboratories consists of, first, making twofold dilutions of virus in 0.25 cc. amounts ranging from 1:5 to 1:1,280 in a series of tubes; second, adding 0.25 cc. of 1:5 saline dilutions of serum to each tube; and third, adding 0.25 cc. of a 0.5 per cent suspension of red cells to each tube. The appearance of the pattern formed by the sedimented cells is shown in Figure 21.4.

The hemagglutination test is a reliable means of differentiating embryo-propagated pneumoencephalitis virus from the viruses of clinically similar infectious bronchitis and laryngotracheitis since the allantoic fluid of embryos infected with the latter two viruses have been shown to be incapable of causing red cell agglutination.

The HI test is now regarded as a highly reliable means of identifying pneumoencephalitis, provided the inhibiting titer of the serum is found to be 80 or higher. HI titers of 10, 20, or 40 must at present be considered as of questionable diagnostic significance because in many instances evidence of pneumoencephalitis in the donors of serum with such HI titers could not be obtained by neutralization tests or challenge of the birds with

DILUTIONS OF SERUM

| 1/10 | 1/20 | 1/40 | 1/80 | 1/160 | 1/320 | 1/640 | 1/1280 | 1/2560 |



1    2    3    4    5    6    7    8    9
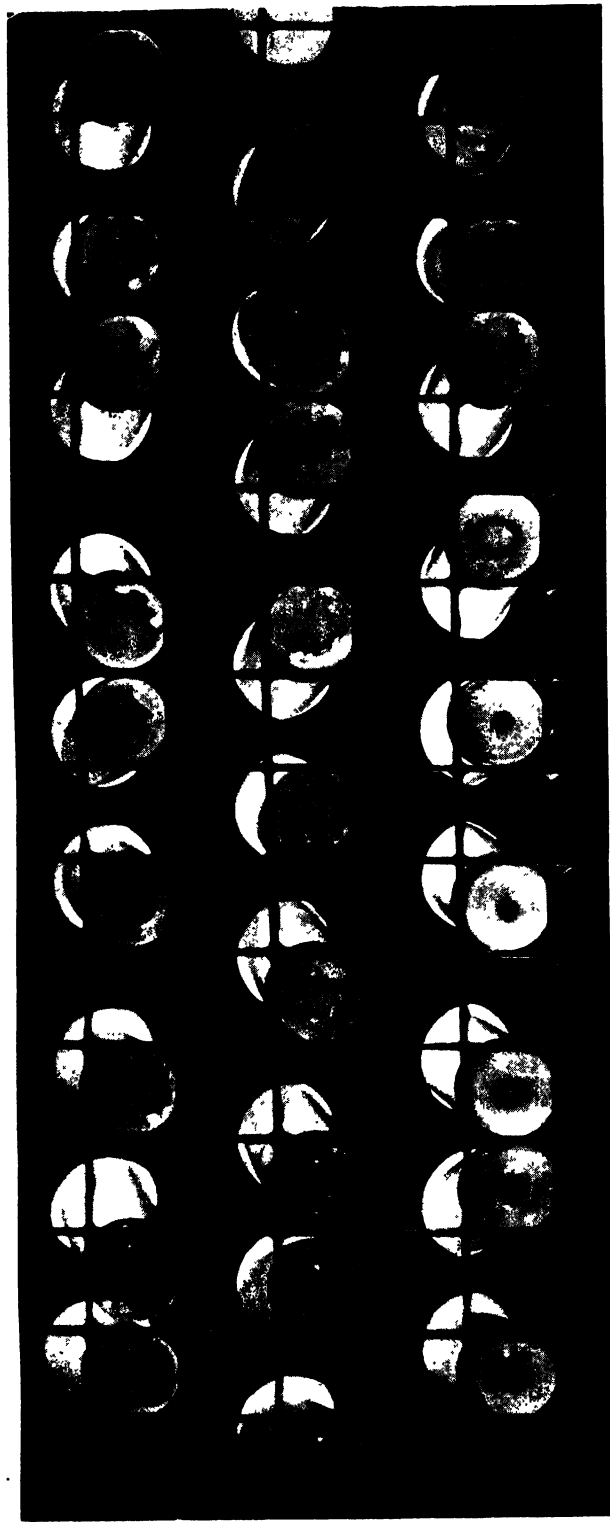
Fig. 21.4. The hemagglutination-inhibition test with two negative serums (top and middle row) and one positive (bottom row) serum. Each tube contains 0.25 cc. of a 1:80 dilution of virus, 0.25 cc. of serum diluted as indicated above, and 0.5 cc. of the red blood cell suspension. All tubes in the top and middle rows show ++++ agglutination of red cells which indicates that no HI antibodies were present in the serums. In the bottom row, tubes 1–5 show no agglutination of red cells, tube 6 ++ agglutination, and tubes 7–9 ++++ agglutination of red cells. This indicates that the amounts of serum in tubes 1–6 inhibited hemagglutination. The HI titer of the serum is expressed as 1:160. The irregular outlines of the blanket of cells on the bottom of many of the tubes showing ++++ hemagglutination resulted from the agglutinated cells starting to slide toward the center. Photo taken 30 minutes after test was set up. (Read dilutions from the left.)

virus. The reason for low inhibiting power of serum, in some instances, may be that the infection of the birds is too recent for higher concentration of HI antibodies to have developed in their serum. Therefore, when the HI test reactions of serum from a suspected flock are only strong enough to be reported as suspicious, additional serum samples for testing should be obtained.

Blood samples for the HI test should, preferably, be drawn aseptically from the wing vein or heart with a sterile hypodermic syringe and transferred to a sterile, tightly stoppered vial. Any method, however, by which blood samples are being secured and delivered to a laboratory in suitable condition for the pullorum test would suffice. In fact, blood samples taken for the pullorum test could also be used for the HI test in making a survey of the distribution of pneumoencephalitis. When blood samples cannot be delivered to a laboratory for the HI test within 24 hours after they are drawn, or if they will be exposed to a high temperature in transit, the serum only should be sent. If desired, the serum of two or more birds from the same flock can be pooled. The addition of one or two drops of chloroform to each cc. of serum is advisable to inhibit bacterial growth.

**Neutralization test.** This test has been in use for many years for identifying viruses and establishing immunological relationship between them. It is based on the ability of antibodies in the serum of animals immune to any one of several virus diseases, including pneumoencephalitis, to render noninfective or neutralize the viral causative agent when the two are mixed together in a test tube; i.e., *in vitro,* in correct proportions. The test is conducted in such a way that it is both qualitative and quantitative, in that it both determines the presence in the serum of antibodies for a particular virus and measures their virus-neutralizing power. A procedure for utilizing the test in the diagnosis of pneumoencephalitis is as follows: Equal parts of serum from a recovered suspected case and serial tenfold saline dilutions of virus; i.e., allantoic fluid of an infected embryo, are mixed in a series of test tubes. A like series of serum-virus mixtures using normal serum and a series of tenfold saline dilutions of virus alone carried beyond the highest dilution which is capable of producing infection in embryos or chicks are also made. At least three embryos or two chicks are then inoculated with a uniform dose of each serum-virus mixture and of each dilution of virus. The results of inoculation with the virus alone determine the infective titer of the virus, the amount of virus in the highest dilution which proves infective being termed one m.i.d. (minimum infective dose) . Neither the normal serum nor serum from a bird which has had any disease other than pneumoencephalitis should influence the infectivity of the virus. On the other hand, the serum of immune birds neutralizes the virus so that part or all of the mixtures of the two are not infective. For example, the virus that is

infective in all tenfold dilutions up to 1:1,000,000 $(10^{-6})$ may not infect in dilutions higher than 1:100 $(10^{-2})$ when an equal amount of undiluted immune serum is added to each virus dilution. This would show that a volume of virus; i.e., allantoic fluid of an infected embryo, containing 10,000 infective doses (m.i.d.) of virus was not infective when it was mixed with an equal quantity of the serum and, therefore, that the donor of the serum had been infected with pneumoencephalitis. The neutralizing titer would be expressed as $10^4$. Serum for the neutralization test should be procured as described for the HI test.

**Immunity tests of live birds.** In selecting birds to be submitted to a laboratory for this test one should be certain that they became infected at least one to two weeks previously so that there has been time for them to have become immune. The test, of course, consists simply of inoculating these birds and normal controls with a challenge dose of virus. It is customary to use previously titrated virus so that the suspects are challenged with a known dose. Resistance or susceptibility of the suspected birds to challenge indicates, respectively, whether the disease with which they had been affected was or was not pneumoencephalitis. An advantage of submitting live birds instead of their serum only is that both the serological tests and challenge can, if desired, be employed in making the diagnosis.

**Control and prevention.** There does not appear to be any measure of proved effectiveness for preventing the spread of the disease from the initially infected pen of birds to other susceptible groups on a farm, or of modifying the severity of the disease with respect to mortality and adverse effect on the development of the young and the egg yield of layers. By careful observance of precautions against carrying the infection from one unit to another, it may be possible to prevent the immediate spread from infected to other well separated ones, but one cannot be assured that this procedure will accomplish more than postponement of the appearance of the disease in the other units. Brooder chicks, for example, may escape infection present among adult birds only to become infected later in life. The sacrifice of the unit in which the disease is first recognized, provided it is detected early and the infection has not yet reached other units, would terminate an outbreak. The aforementioned difficulty in identifying and detecting the disease in its early stages, however, makes it hard to decide when it would be propitious to adopt this procedure. Good care is the only thing that can be suggested in the way of treatment for an affected flock. Ample warmth should be provided and efforts made to correct the decreased food intake which results from the impaired appetite. The latter may be accomplished in some measure by changes in the feeding practice such as giving mash in meals instead of keeping it constantly before the birds, supplying additional milk products, and daily extra feeding of rolled barley soaked in milk, moistened mash, and

fresh tender greens. This may serve to shorten the period of convalescence following an outbreak in either young or adult chickens.

Observation has shown that the disease is likely, although not certain, to recur annually during the years following an initial outbreak on a farm. Whether the origin of the infection during the first and subsequent years following, however, was from residual infection in virus carriers among the previously infected birds or from reintroduction of virus from neighboring farms or other outside sources has not been determined in most instances. Presumptive evidence incriminating virus carriers as a possible common source is found in the fact that the carrier state in recovered birds has been demonstrated (Beach, 1942b). When visual evidence of the infection is lacking it is well to have the flock checked by serological tests to make certain that the disease has not occurred in the subclinical form.

Some precautions that may assist in preventing recurrence of the disease on a farm from residual infection among recovered birds and also in avoiding introduction of virus to incite either initial or recurrent outbreaks are as follows: (1) Sell all layers for slaughter at the end of the first laying year. Keep the young replacement stock well segregated until all layers are removed and permanently segregated from any of the older birds which are retained as breeders. There is a reasonably good chance that the disease may be eradicated permanently or for a period of years by the conscientious use of this procedure. Serious consideration should be given to complete depopulation before the replacement chicks are secured, in case the disease occurs in the poultry on a farm which is well removed from flocks on other farms. This plan is an official eradication measure in at least one state. (2) Thoroughly clean and disinfect all houses and equipment used by the layers well in advance of the time they will be used for the replacement stock. (3) Bring no poultry onto a farm except as day-old chicks (or poults). In this connection it should be pointed out that the disease has been introduced into flocks previously pneumoencephalitis-free by started chicks and young breeding males. (4) Eggs from or flocks which have had pneumoencephalitis should not be used for hatching until at least 30 days after the production rate equals or exceeds the preinfection rate. A poultry raiser should ascertain that the hatchery which supplies his chicks rigidly adheres to this requirement. (5) Do not bring onto a farm feed sacks or other articles which have been in other poultry establishments of any nature or otherwise been in contact with poultry, and which, in the meantime, have not been properly cleaned and disinfected by heat or chemical means. (6) Allow no visitors inside poultry houses or yards. (7) Do not permit part-time laborers, blood-testers, flock inspectors, or other persons who must work inside poultry houses, to wear footwear or other outer clothing which has not been properly cleaned and disinfected since last worn in poultry establishments of any

nature, unless it is certain that such other establishments are free from pneumoencephalitis infection.

Because of the apparent ease with which infection can spread from infected flocks to nearby farms, the effectiveness of these precautionary measures in keeping a flock free from the disease is likely to be less in a congested poultry district than in areas in which the flocks are scattered and well separated.

**Vaccination.** Experiments in the development of formalin-inactivated vaccine from chicken embryos for use in the control of pneumoencephalitis were undertaken at the University of California in 1942 (Beach, 1942b). After data favorable to vaccination had been accumulated by laboratory experiments, in which the antigenicity of vaccines was tested by challenging vaccinated birds by inoculation with cultured virus, extensive controlled field trials were undertaken and have been continued through each succeeding year up to the present. The results of the controlled field trials are discussed later. Promising results of similarly conducted laboratory experiments with formalin-inactivated embryo vaccine were reported by Brandly and co-workers (1946b) in 1946. These investigators stated having found that the immunogenic response to formalin-killed virus was augmented and prolonged by the addition of certain "adjuvant" substances. They also made the interesting observation that the antigenicity of vaccine was reduced by the inclusion of the yolk of eggs laid by immune hens.

Beach (1944b) reported that in field trials the vaccine did not produce the degree of protection against natural infection that had been expected from the results of the laboratory experiments. Subsequent field trials with formalin-inactivated vaccines of varying constitution to some of which "adjuvant" substances were added showed no greater effectiveness (Beach and DeLay, 1947, unpublished data).

In reporting the results of large-scale controlled vaccination field trials, Beach (1944b) stated that the vaccinated portions of the flocks did not exhibit complete protection against natural infection; but uniformly, they were appreciably less severely affected by the disease than the nonvaccinated controls with respect to both mortality and loss from decreased egg yield. The effect of the disease on egg yield was said to have been lessened to the extent that vaccination was worthwhile from that standpoint alone. The accumulated data (unpublished) from subsequent controlled field vaccination trials conducted by Beach and DeLay show that the antigenic activity of these vaccines against natural infection was no greater than had been obtained in the earlier field trials and was not enhanced by the inclusion of "adjuvant" substances in the vaccine. It was also demonstrated that single doses of vaccine were less effective than an equal quantity given in two doses, and that the interval between the two doses could be varied from a week to as

long as three months without influencing the effectiveness of the vaccination. Commercially produced vaccine became available in 1945. This has been used on a large number of chickens but has not been adopted by poultrymen for routine use as has chicken pox and laryngotracheitis vaccines. The reasons for this are believed to be the relatively high cost of the vaccine, inability of the vaccine to confer complete immunity, and the relatively small loss by death which poultrymen have experienced from outbreaks of the disease in their flocks. The results of field use of formalized vaccine during a period of more than five years do not make it appear probable that a vaccine of this type will be developed which will solidly immunize chickens against the disease. Therefore, it is improbable that it will be employed for eradication of the disease on an area or nationwide basis by universal vaccination.

The failure of vaccine containing chemically inactivated virus to give full protection against natural infection has stimulated experiment station workers and research staffs of commercial laboratories to search for a naturally occurring virus of sufficiently low virulence or for artificial means whereby the virulence of virus can be sufficiently modified so that a safe and effective live-virus pneumoencephalitis vaccine can be made available. It is known that encouraging progress has been made at several laboratories, but, thus far, only one (Van Roekel et al., 1948) published account of this work has appeared. These investigators reported having obtained "strains of virus of low virulence which can be used by the stick method (web of wing) to immunize sexually immature birds without producing an active outbreak of the disease." They pointed out, however, that the virus caused a systemic reaction manifested by slight depression and inappetence and an adverse effect on egg production and egg quality as is the case in a natural outbreak.

### REFERENCES

Beach, J. R.: 1940. A nervous disorder of chicks. Nulaid News 18:13.

——: 1942a. A respiratory-nervous disorder of chickens. Nulaid News 20:9.

——: 1942b. Avian pneumoencephalitis. Proc. 46th Ann Meet. U. S. Livestock Sanitary Assn., p. 203.

——: 1944a. The neutralization in vitro of avian pneumoencephalitis virus by Newcastle disease immune serum. Science 100:361.

——: 1944b. Vaccination for pneumoencephalitis. Proc. 48th Ann. Meet. U. S. Livestock Sanitary Assn., p. 177.

——: 1947. The application of the hemagglutination-inhibition test in the diagnosis of avian pneumoencephalitis. Jour. Am. Vet. Med. Assn. In Press.

Beaudette, F. R.: 1943. A review of the literature on Newcastle disease. Proc. 47th Ann. Meet. U. S. Livestock Sanitary Assn., p. 122.

——: 1946. Newcastle disease in poultry. Cornell Vet. 36:105.

—— and Black, J. J.: 1945. Newcastle disease in New Jersey. Proc. 49th Ann. Meet. U. S. Livestock Sanitary Assn., p. 49.

Berg, L. R., Bearse, G. E., and Hamilton, C. M.: 1947. The effect of Newcastle disease on egg production and egg quality. Poultry Sci. 26:614.

Brandly, C. A., Moses, H. E., and Jungherr, E.: 1946. Transmission of antiviral activity via the egg and the role of congenital passive immunity to Newcastle disease in chickens. Am. Jour. Vet. Res. 7:333.

Brandly, C. A., Moses, H. E., Jungherr, E., and Jones, E. E.: 1946a. The isolation and identification of Newcastle disease virus. Am. Jour. Vet. Res. 7:289.

————, Moses, H. E., Jungherr, E., and Jones, E. E.: 1946b. Immunization of chickens against Newcastle disease. Am. Jour. Vet. Res. 7:307.

Burnet, F. M.: 1942. The affinity of Newcastle disease virus to the influenza group. Australian Jour. Exper. Biol. and Med. 20:81.

DeLay, P. D.: 1947. The isolation of avian pneumoencephalitis virus from the yolk sac of 4-day-old chicks, from embryos, and infertile eggs. Science 106:545.

————, DeOme, K. B., and Bankowski, R. A.: 1947. Recovery of the virus of pneumoencephalitis (Newcastle disease) from the air of a poultry house containing infected birds. Unpublished manuscript.

Dobson, N.: 1939. Newcastle disease. Proc. Seventh World's Poultry Cong., p. 250.

Doyle, T. M.: 1927. A hitherto unrecorded disease of fowls due to a filter-passing virus. Jour. Comp. Path. and Therap. 40:144.

————: 1933. The virus diseases of animals with special reference to those of poultry. Jour. Comp. Path. and Therap. 46:90.

Fenstermacher, R., Pomeroy, B. S., and Malmquist, W. A.: 1946. Newcastle disease in Minnesota. Proc. 50th Ann. Meet. U. S. Livestock Sanitary Assn., p. 151.

Hirst, G. K.: 1941. The agglutination of red cells by allantoic fluid of embryos infected with influenza virus. Science 94:22.

————: 1942. The quantitative determination of influenza virus and antibodies by means of red cell agglutination. Jour. Exper. Med. 75:49.

Jungherr, E.: 1946. Proceedings of conference on Newcastle disease. U. S. Dept. Agr. P. 93.

————: 1948. Report of the committee on modes of spread of Newcastle disease. Jour. Am. Vet. Med. Assn. 112:124.

———— and Terrell, N.: 1946. Observations on the spread of Newcastle disease. Proc. 50th Ann. Meet. U. S. Livestock Sanitary Assn., p. 158.

Levine, P. P., Fabricant, J., and Mitchell, G. B.: 1947. Newcastle disease in ring-necked pheasants. Cornell Vet. 37:265.

Lorenz, F. W., and Newlon, W. E.: 1944. Influence of avian pneumoencephalitis on subsequent egg quality. Poultry Sci. 23:193.

Lush, D.: 1943. The chick red cell agglutination test with the viruses of Newcastle disease and fowl plague. Jour. Comp. Path. and Therap. 53:157.

Minard, E. L., and Jungherr, E.: 1944. Neutralization tests with pneumoencephalitis virus. Am. Jour. Vet. Res. 5:154.

Reagan, R. L., Lillie, M. G., Poelma, L. J., and Brueckner, A. L.: 1947a. Transmission of the virus of Newcastle disease to the Syrian hamster. Am. Jour. Vet. Res. 8:136.

————, Lillie, M. G., Poelma, L. J., and Brueckner, A. L.: 1947b. Response of some mammals to Newcastle virus. Am. Jour. Vet. Res. 8:427.

Stafseth, H. J., Beaudette, F. R., Carpenter, C. D., Delaplane, J. P., Jungherr, E., Hendricks, W. H., and Hinshaw, W. R.: 1944. Report of committee on transmissible diseases of poultry. Proc. 48th Ann. Meet. U. S. Livestock Sanitary Assn., p. 197.

Stater, A. E.: 1945. The poultry industry in India. World's Poultry Sci. Jour. 1:46.

Stover, D. E.: 1942a. A respiratory-nervous disorder. Nulaid News 20:12.

————: 1942b. A filtrable virus, the cause of a respiratory-nervous disorder of chickens. Am. Jour. Vet. Res. 3:207.

————: 1942c. Respiratory-nervous disorder in 8-months' old pullets. Am. Jour. Vet. Res. 3:239.

Van Roekel, H.: 1946. Annual report. Mass. Agr. Exper. Sta., Bul. 436:66.

————, Sperling, F. G., Bullis, K. L., and Olesiuk, O. M.: 1948. Immunization of chickens against Newcastle disease. Jour. Am. Vet. Med. Assn. 112:131.

# CHAPTER TWENTY-TWO

# PSITTACOSIS (ORNITHOSIS)

*By* K. F. MEYER, *The George Williams Hooper Foundation for Medical Research, University of California, San Francisco, California*

☆   ☆   ☆

Psittacosis, an apparent but more frequently inapparent infection caused by infective coccoid elementary bodies, *Miyagawanella psittaci*, intermediate between the Rickettsiae and the filtrable viruses, is found in parrots, lorikeets, and cockatoos in Australia, the jungles of South and Central America, and in commercial parakeet breeding establishments of the temperate zone. Aviaries in the United States, Germany, and Argentina harbor parakeets which transmit the disease to canaries, finches, and Java sparrows. Latent infections in pigeons (*Columba livia*), ducks (*Anas platyrhynchos*), and chickens (*Gallus gallus*), are economically and epidemiologically significant. Infections caused by viruses morphologically and biologically indistinguishable from psittacosis virus have been demonstrated in fulmars (*Fulmar glacialis*), American Herring gulls (*Larus argentatus smithsonianus*), and willets (*Catoptrophorus semipalmatus*). Psittacosis-like viruses have been isolated from the respiratory tracts of healthy or diseased mammals (mice, Gönnert and Nigg and Eaton; and cats, Baker).

## HISTORY AND DISTRIBUTION

Ritter, at Ulster, Switzerland, in 1879 described an illness among members of a household as "pneumotyphus," which he attributed to a consignment of tropical birds recently secured from Hamburg. Some of the parrots, finches, and exotic birds were sick when they were received; autopsies on the birds—the type of parrot was not stated—were negative. Shortly afterwards Eberth and Wolff examined gray parrots from the Guinea Coast in an endeavor to explain the immense mortality among consignments to Germany and found micrococci in great numbers in the organs. The malady in man became known generally through the outbreaks of severe pneumonia which occurred in Paris in 1892. At first, Dujardin-Beaumetz concluded that the epidemic was of human origin and not a parrot disease transmissible to man. Peters, and later Dubief (January, 1893), through study of a new epidemic concluded that the disease was a specific illness conditioned by contact with an infected parrot. This hypothesis received strong support

from the studies of Nocard (1892) who cultivated a Gram-negative, motile bacillus from the bone marrow of the dried wings of parrots which had died on the voyage from Buenos Aires some four months before. This bacillus was pathogenic for fowls, mice, and guinea pigs as well as for parrots, both by inoculation and feeding. Nicolle, in 1898, and subsequent investigators, however, failed to obtain proof that psittacosis [from psittacus = parrot, so designated by Morange (1895)] of parrots and man was caused by Nocard's bacillus.

Between 1894 and 1897, outbreaks of the disease occurred in Italy and were traced to recent importations of Amazon parrots from Buenos Aires. Although psittacosis associated with a shipment of parakeets, held in a box at a post office building at Dühringsdorf near Landsberg, Germany, resulted in eight human infections, the first big epidemics occurred in Cologne in 1898 and in Krefeld in 1899. Imported green Amazon and gray parrots were held responsible. Leichtenstern regarded it as probable, but not susceptible to proof, that contagion from the sick parrots had occurred. He was greatly impressed by the human case-to-case infections. In the famous Zülpich epidemic in the spring of 1909, two apparently healthy parakeets infected twenty-six persons who entered the room where the birds were held. Bacteriological examination of the killed parakeets yielded slightly hemolyzing streptococci, but Nocard's bacillus was not found. Single and group infections have been reported sporadically between 1914 and 1928 from the United States and England. The epidemic of 1917 reported by McClintock originated in the basement of a large department store where many sick parrots were held and sold. Store employees and purchasers of the birds became ill, and the association between human disease and sick parrots was quite obvious. Jackson, Hull, and Rucker, however, claimed the epidemic to be an unusually severe influenza and offered as proof negative bacteriological findings in man and parrots. Many coccoid and Gram-negative bacillary forms were isolated by McClintock from his autopsies of parrots. On passage through mice the bacilli became Gram-negative, slender, club-shaped, motile bacilli. Thus, no light was thrown on the etiologic factors of psittacosis until the widespread outbreak of 1929–30.

During the summer and fall of 1929, Barros (1940) informed a number of prominent physicians and later the Medical Society of Córdoba, Argentina, of over 100 cases of a serious and peculiar pneumonia among inhabitants in the province. He diagnosed the infection as psittacosis because epidemiologic investigations showed that a consignment of 5,000 psittacine birds, imported into Argentina from Brazil, and offered for auction under the most insanitary conditions, served as the focal center for dissemination of the illness. A destructive infection (in the light of present-day knowledge, unquestionably psittacosis) had broken out, and the managers, anxious to

sell as many living birds as possible, disposed of their stocks with great rapidity. Purchasers and re-purchasers, auctioneers, etc., became ill, and some died. The auction then was transferred to Tucuman; birds continued to die, and human cases flared up in every quarter of the city. Local attention was directed to the strange disease when several epidemics developed in the capitol of Argentina, Buenos Aires, during the month of October. There were two tragic deaths among twelve members of a theatrical troupe, all of whom fell ill following the use on the stage of a parrot which came from the original importation to Córdoba. These events fully warned the population, and trade in parrots was stopped entirely in Argentina. Passengers of steamers calling at the ports, however, ignorant of the epidemic, bought many infected birds from unscrupulous dealers. The malady was conveyed to at least twelve different countries. It reached the United States in November, while cases already had been reported in England in July and were reported there again in December. During the early months of 1930, newspapers gave accounts in Austria, Italy, Switzerland, France, Denmark, Algeria, Holland, Egypt, Czechoslovakia, Germany, and Sweden. It was stated in many of the reports that shipments of sick parrots had arrived before cases were observed. A critical perusal of the records of 1930, however, leaves no doubt that the South American parrots were not the only sources of infection. General interest in the new disease called attention to these infections, and what, under ordinary circumstances, would have been dismissed as atypical pneumonia was correctly diagnosed as psittacosis. In England, the United States, Austria, and Switzerland, parrot fever was observed to develop following contact with love-birds and canaries. The pandemic era of 1929–30 with approximately 750 or 800 human cases of psittacosis was terminated abruptly when, in the early months of 1930, the public health agencies of every affected country established stringent import regulations or quarantine restrictions prohibiting the admission of birds belonging to the large group known as the "Psittaciformes."

Throughout the pandemic, all observers were struck by the negative serological and bacteriological results, both in their patients and in the guilty parrots. A number of investigators soon found that the infective material, when passed through Chamberland filters, was still capable of infecting parrots. Extensive experimental studies by Rivers and associates in the United States and by Bedson in England fully proved and firmly established the filtrable nature of the cause of psittacosis.

The discovery by Meyer and Eddie in 1931 and 1932 of a wide distribution of latent psittacosis in the local breeding establishments and aviaries of California, and in Germany by Fortner and Pfaffenberg, in Austria by Gerlach, in France by Aujaleu and Jude (1936), in Holland by Ruys (1934), in England by Levinthal, and in Canada by MacNabb in 1941, offered a

## TABLE 1

### INCIDENCE OF HUMAN PSITTACOSIS IN DIFFERENT COUNTRIES
(Contact with Psittacine Birds)

| Country | Period 1876–1928 | | | | Pandemic 1929–30 | | | | Period 1931–46 | | | | Total | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Parrots | | Parakeets | | Parrots | | Parakeets | | Parrots | | Parakeets | | | |
| | Cases | Deaths | Cases | Deaths | Cases | Deaths | Cases | Deaths | Cases | Deaths | Cases | Deaths | Cases | Deaths |
| United States | 4(++) | — | | | 165 | 32 | 4 | 1 | 6 | 3 | 317 | 40 | 496 | 76 |
| Honolulu | — | — | | | 2 | 1 | — | — | — | — | — | — | 2 | 1 |
| Canada | — | — | | | — | — | 7 | — | — | — | 43 | 1 | 50 | 1 |
| Argentina | — | — | | | 100 | 13 | — | — | 11 | 3 | 56 | 16 | 167 | 32 |
| Brazil | 1 | — | | | +? | + | — | — | — | — | — | — | 1+ | + |
| Algeria | .1 | — | | | 11 | 5 | — | — | 10 | — | — | — | 22 | 5 |
| Australia | — | — | | | — | — | 11 | — | — | — | — | — | 12 | — |
| Austria | — | — | | | 7 | 0 | 7 | 1 | — | — | 18 | 1 | 32 | 2 |
| Czechoslovakia | — | — | | | 6 | — | — | — | — | — | — | — | 6 | — |
| Denmark | — | — | | | 10 | — | — | — | 4 | — | — | — | 14 | — |
| Faroe Islands | — | — | | | — | — | — | — | — | — | — | — | — | — |
| Iceland | — | — | | | — | — | — | — | — | — | — | — | — | — |
| Egypt | — | — | | | 1 | 1 | — | — | — | — | — | — | 1 | 1 |
| France | 77 | 28 | | | 20 | 1 | — | — | — | — | 9 | 1 | 106 | 30 |
| Germany | 62 | 9 | 45 | 1 | 215 | 45 | 3 | 1 | — | — | 439 | 47 | 761 | 102 |
| Great Britain | 7 | 3 | | | 117 | 24 | — | — | — | — | 8 | — | 135 | 28 |
| Italy | 20 | 12 | | | 5 | 2 | — | — | — | — | — | — | 25 | 14 |
| Japan | — | — | | | 1 | — | — | — | — | — | — | — | 1 | — |
| Mexico | — | — | | | 1 | 1 | — | — | — | — | — | — | 1 | 1 |
| Netherlands | — | — | | | 9 | 3 | — | — | 5 | — | 16 | 3 | 30 | 6 |
| Poland | — | — | | | 2 | — | — | — | — | — | — | — | 2 | — |
| Spain | — | — | | | + | + | — | — | — | — | — | — | + | + |
| Sweden | — | — | | | 6 | 1 | — | — | — | — | — | — | 6 | 1 |
| Switzerland | — | — | | | 46 | 3 | 7 | — | — | — | 2 | — | 55 | 3 |
| Total | 171 | 52 | 45 | 1 | 724 | 131 | 39 | 3 | 36 | 6 | 910 | 109 | 1,925 | 302 |

TABLE 1—*Continued*

INCIDENCE OF HUMAN PSITTACOSIS IN DIFFERENT COUNTRIES

(Contact other than Psittacine Birds)

Period 1931–46

| Country | Pigeons | | Chickens | | Canaries | | Fulmars | | Ducks | |
|---|---|---|---|---|---|---|---|---|---|---|
| | Cases | Deaths | Cases | Deaths | Cases | Deaths | Cases | Deaths | Cases | Deaths |
| United States | 103 | 7 | 4 | 1 | 12 | 1 | .... | .... | 5 | 0 |
| Argentina | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... |
| Faroe Islands | .... | .... | .... | .... | .... | .... | 186 | 38 | .... | .... |
| Iceland | .... | .... | .... | .... | .... | .... | 5 | ? | .... | .... |
| Germany | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... |
| Netherlands | .... | .... | .... | .... | 1 | 0 | .... | .... | .... | .... |
| Total | 103 | 7 | 4 | 1 | 13 | 1 | 191 | 38 | 5 | 0 |

Period 1931–46

| Country | Grouse | | Multiple* Exposures | | Laboratory Infections | | Human to Human | | Miscellaneous | | Total | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Cases | Deaths | Cases | Deaths | Cases | Deaths | Cases | Deaths | Cases | Deaths | Cases | Deaths |
| United States | 2 | 0 | 10 | 0 | 12 | 0 | 11 | 2 | 68 | 8 | 227 | 19 |
| Argentina | .... | .... | 12 | 6 | 3 | 1 | 4 | — | .... | .... | 19 | 7 |
| Faroe Islands | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 186 | 38 |
| Iceland | .... | .... | .... | .... | 3 | 1 | .... | .... | .... | .... | 5 | ? |
| Germany | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 3 | 1 |
| Netherlands | .... | .... | .... | .... | .... | .... | .... | .... | .... | .... | 1 | 0 |
| Total | 2 | 0 | 22 | 6 | 18 | 2 | 15 | 2 | 68 | 8 | 441 | 65 |

* Two or more species of birds.

splendid opportunity for thorough study of the disease from clinical, etiological, and epidemiological points of view. Once the clinical picture of psittacosis had become familiar and laboratory methods for its diagnosis had been developed, further cases not associated with South American parrots or parakeets continued to be recognized. When Meyer and Eddie found several infected birds in a cargo of native Australian budgerigars on its arrival in California (1934a), although there had been no contact with any known source of infection, and when two consignments of Australian parrots heavily infected with psittacosis arrived in London (Levinthal, 1935), Burnet initiated his detailed investigations on psittacosis in wild Australian parrots. The disease was found in the true parrots, the lorikeets, and the cockatoos. The parrot-to-man infection-chain thus appeared to constitute the sole problem with the order of PSITTACIFORMES as the principal reservoir of infection, while certain highly susceptible species of finches and canaries became diseased merely through contact with diseased parrots or parakeets. This belief soon was recognized to be ill-founded when Haagen and Mauer (1938) showed the fulmar or petrel in the Faroe Islands (*Fulmarus glacialis*) to be infected and to be the source of human infection.

## DISTRIBUTION AND PREVALENCE

Further knowledge has altered the concepts of the avian disease. Studies by Meyer, Eddie, and Yanamura (1942), Smadel, Wall, and Gregg (1943), Andrewes and Mills (1943), and Labzoffsky (1947) established its worldwide distribution in wild and domesticated pigeons. Recently, demonstration of spontaneous infections in duck-breeding establishments in California and New York (Eddie and Meyer, 1946), and in seashore birds (Eddie and Meyer, 1946; Pollard, 1947) has proved its wide distribution in the bird kingdom.

Official reports, published accounts of house epidemics, and the records of the Hooper Foundation dealing with human psittacosis have been summarized in Table 1. The world-wide distribution is evident. The statistical data, however, must be considered approximations which deal with frank clinical cases; no attempt has been made to secure information concerning the subclinical or mild cases, often suspected but only occasionally proved serologically, which are encountered with increasing frequency in recent years. The persistently high incidence of human psittacosis in Germany and the United States is with few exceptions attributable to the distribution and sale of locally bred and raised parakeets. It is important to note, however, that with the progress of control measures against diseased birds, the number of cases of human psittacosis annually reported in the United States declined to three in 1937 and 1938. It rose to nineteen in 1939 and was seventeen in

1940, fifty in 1941, and eighty-six in 1942. It dropped to twenty-one in 1943 and to thirty in 1944, but rose again to ninety-six in 1945; fifty-three cases were reported in 1946.

## GENERAL EPIZOOTIOLOGY AND EPIDEMIOLOGY

Enzootic psittacosis among parrots, parakeets, or pigeons takes the form of a latent inapparent infection, producing no visible symptoms and no pathological signs beyond an enlarged spleen. Many of the birds become infected in the nests, and remain immune to reinfection from other sources. Variations from this condition may favor either host or parasite. High mortalities in importations or zoological gardens (London, Washington, Melbourne) are due either to the fact that the parrots escaped nest infections, or that disturbances in the environment (low temperature, crowding in insanitary cages, improper feeding) induced relapses. Virus dispersed in large quantities and in crowded quarters spreads the infection rapidly from diseased to susceptible birds. A variable number may die, but a great many carriers develop. Under these circumstances, it is not surprising if birds apparently healthy when captured in the jungle or bush or housed under the usual conditions are found to be suffering from psittacosis by the time they reach their ultimate purchasers.

Burnet has observed outbreaks of fatal psittacosis in wild King parrots (*Alisterus* sc. *scapularis*) in the hilly, timbered country of Victoria, in rosellas (*Platycercus eximius*) in Tasmania, and among parrots in the southeastern district of South Australia. Existence of the endemic disease among paroquets (*Myiopsitta monachus* Boddaert) in the Province of Salta, North Argentina, was recently proved by Parodi and Silvetti (1946). Dead paroquets were observed in the trees; of 150 caught only 85 survived transportation to Tucuman, where they initiated eight cases of human psittacosis with two deaths. Two parrots of the original lot and one of four birds caught in the wild were proved to be infected.

Either fleeting or prolonged exposure in a room, house, pet store, or aviary where visibly diseased or apparently healthy infected parrots, parakeets, canaries, and pigeons are held in captivity may result in human infections. Usually, the birds have been recently acquired. A single case in the past could escape detection, but in the past few years the complement-fixation test has been of great assistance in proving these atypical, influenza-like forms of pneumonia or pneumonitis to be psittacosis. Quite often in rapid succession, additional illnesses among relatives and even guests and visitors of the infected person create the well-known house epidemics which give the epidemiology of psittacosis its characteristic, rather stereotyped pattern. Epidemiologic records show that the cleaning of petrels, pigeons, ducks, and

chickens causes human psittacosis. In rare instances, handling of feathers has resulted in infection. Usually two to three weeks elapse between the acquisition of the birds and onset of the first case. A seasonal prevalence during the winter months (January–April) probably is due to the frequency with which human beings receiving birds as gifts are brought in contact with them in the closed rooms of a winter household. The great epidemics of the past occurred during the winter months, and in Germany and Argentina the predominance of the disease during the colder months is striking. It is well to remember, however, that severe psittacosis on the Faroe Islands and among pigeon fanciers is not uncommon in midsummer or early fall. The majority of psittacine infections have occurred in people of middle age; children under ten years of age have a low susceptibility (in England, among 104 cases, only four were under ten years; in Germany, among 160 of known age, only two were under ten; in California, among 92 cases, one was a boy $8\frac{1}{2}$ years old). Aside from the low incidence, it is a common experience that infection in children is much milder than in the older patients in the family. The greater frequency in females [California, 60 women : 31 men; Germany, 33 women : 19 men (Pfaffenberg), and 17 female fulmars : 8 male fulmars (Haagen and Mauer)] is in part due to the fact that women more often than men are engaged in the breeding of parakeets or the preserving of fulmars, or that as lovers of pets they more frequently come in contact with birds. Where the interest in bird-raising has shifted to the male sex, their ability to become infected is well documented by an increased incidence. An infection which in the avian and mammalian host so frequently produces latent subclinical infection, in all probability induces a similar state in man.

A convalescent human carrier with a serum complement-fixation titer of 1:256 whose sputum contains the virus eight years after a laboratory infection with parakeet virus has been reported by Meyer and Eddie (1947).

The *case-fatality rate* of the reported cases has been remarkably uniform both in the United States and in Germany. For 92 cases in California, it is 22 per cent. According to Fortner and Pfaffenberg (1934, 1935), the rate in Germany was 18 per cent in 1933–34, then 20 per cent, and rose to 36 per cent in 1937–38 (Haagen and Mauer, 1939). Among the 186 cases of psittacosis caused by contact with fulmars, 38 (or 20 per cent) died, and among the 15 patients exposed solely to pigeons, 2 (or 13 per cent) died.

Recognition of mild and subclinical infections and introduction of penicillin as a therapeutic agent has reduced the fatality rate; it was 6.0 per cent among 352 cases reported in the United States 1940 to May, 1947. A high mortality, eight deaths in nineteen cases of psittacotic pneumonitis among nursing contacts in an epidemic in the Bayou region of Louisiana, was observed by Olson and Treuting (1944).

Until relatively recently, bird breeders, owners of pet shops, aviculturists,

pigeon raisers, lovers of birds in general, and even veterinarians have doubted the existence of the disease. Until the identical infective agent was demonstrated in the blood and sputum of patients and in the organs of psittacine birds, the diagnosis of parrot fever and the mutual relationship between the apparently healthy parakeet and the "typhoid" pneumonia of its owner were subject to incorrect interpretations by the prejudiced laity. The fact that persons engaged in the breeding, raising, transportation, and sale of psittacine birds are particularly liable to psittacosis was not accepted, though published records amply attested to its verity. The occurrence of psittacosis among bird breeders has been reported by Widowitz (Graz, 1929), Roch and Wohlers (Geneva, 1929), Prausnitz and Stepp (Breslau, 1932), Gerlach (1936b), and MacNabb (Canada, 1940–41); among dealers in parrots, by Marion and Dubois (1892), Barros (1929), Brauer (1930); and among owners of pet shops and department stores, employees in zoological gardens (London and Washington), and finally seamen and baggage car employees connected with the transportation of birds, by Wagner (1886), McClintock, Badger (1929), Ellicot and Halliday (1931), and many others (see Meyer, 1934). Notwithstanding all of these reports, those associated with the shell parakeet trade in California claimed absolute immunity against this malady. They argued that the alleged disease would have attacked primarily those who are intimately in contact with the infected birds, and hence they must be immune. Some credence was given to these arguments until examinations of the sputum and more recently serum-tests proved the so-called "severe attacks of influenza with pneumonia" in bird breeders to be psittacosis.

Of the 114 cases of psittacosis reported in California, 43 (with 4 deaths) or nearly 40 per cent of the total were in owners of parakeet aviaries and pigeon lofts, or members of their families. It is naturally a matter of conjecture, but the few data thus far available from serological tests among raisers of birds would indicate that subclinical infections may explain in part the apparent immunity of some of the men and women who were exposed without any ill effects to heavily infected parakeets, parrots, or pigeons. For example, the serum of a caretaker in one of the infected aviaries gave a complement-fixation reaction in a dilution of 1:16; the serum of an owner of a pigeon loft reacted in a like dilution and at least two of the five keepers of birds, who were exposed to the same sources responsible for two clinical infections in a zoological garden, had antibodies in their sera. Thus, it appears that the supposed immunity in a group of bird breeders either is nonexistent or only relative. One man developed psittacosis about three weeks after acquiring his stock of birds, while five men maintained aviaries with parakeets between two and three years before they became infected. The possibility that the aviaries originally were free from psittacosis and were infected subsequently by purchase, exchange, or barter of diseased shell parakeets cannot

# EPIDEMIOLOGY
# OF PSITTACOSIS AND ORNITHOSIS
## HOUSE EPIDEMICS, OCCUPATIONAL AND ACCIDENTAL INFECTION

Cackatoo

Macaws

Cockatiel

Parrakeets

Germinative Transmission

Amazon Parrot

Masked Love Bird
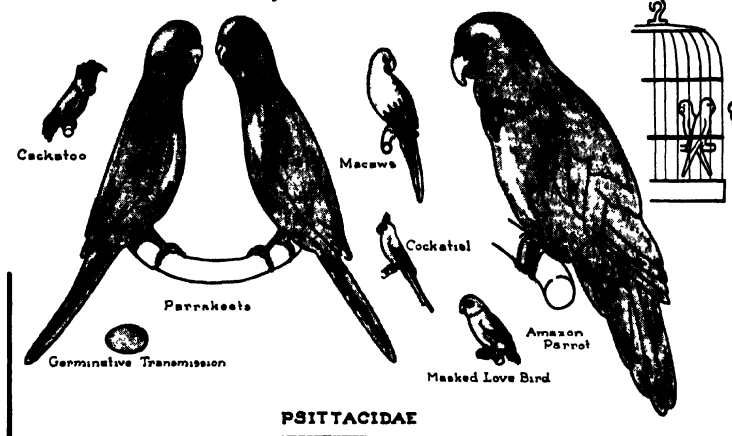
PSITTACIDAE

Miyagawanella
psittaci
(Filterable)
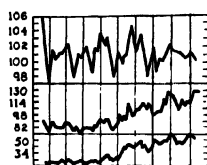
Incubation time - 7-31 days

High temperature with slight remissions.

Toxemia entirely out of proportion to physical findings.

Non productive pneumonia or pneumonitis

Fulmar

PROCELLARIIDAE

Dove

COLUMBIDAE

Pigeon

Gouldian Finch

Zebra Bird   Butterfly Finch   Java Rice Bird

GEOSPIZIDAE

Canary
FRINGILLIDAE
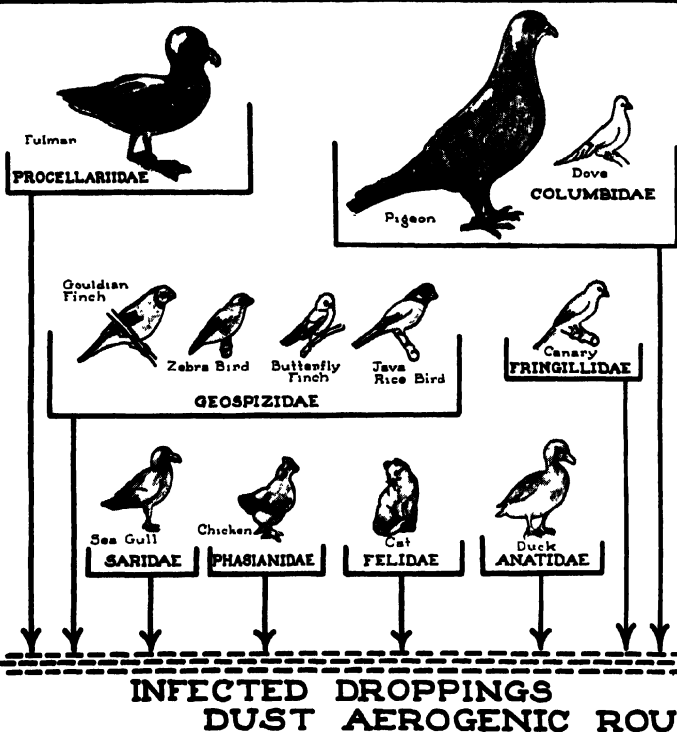
Sputum — Droplet or contact.

Sea Gull   Chicken   Cat   Duck

SARIDAE   PHASIANIDAE   FELIDAE   ANATIDAE

INFECTED DROPPINGS
DUST AEROGENIC ROUTE

Size of birds in relation to incidence and importance.

Experimentally infected: Mice, Guinea Pigs, Hamsters, Pocket Gophers, Rabbits, Monkeys.
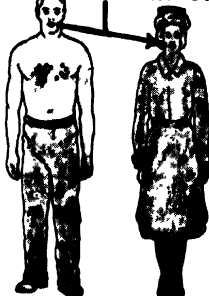
Blood virulent -1st—3rd day.

Sputum virulent
   5th — 23rd day.

Regularly in fatal cases.

Autopsy; virus in lung, spleen and liver.

Complement fixation of serum :—
positive 8th—15th day.

FIG. 22.1. Epidemiology of psittacosis and ornithosis

be excluded in every instance (Hoge, 1934). The heavy exposure in pet shops may lead to a rate of attack ranging from 40 to 50 per cent.

During the height of experimental investigations on psittacosis in 1929 and 1930, thirty-eight laboratory infections were contracted, with five deaths. Despite precautionary measures, seventeen additional cases with two deaths have occurred in the United States, Germany, and Argentina since 1934.

Leichtenstern (1899) observed human-to-human infections, which modern workers have confirmed. At least twenty-three instances involving thirty nurses are known in which contact with sick birds was definitely excluded. Twice repeated human-to-human passage has been observed by Haagen and Krückeberg (1937). Chain transmissions in the third generation have been reported by Hamel. More disconcerting are the transmissions which occur in hospitals. An epidemic in Buenos Aires reported by Loizaga and Averbach (1945) involved twenty-six cases and thirteen deaths, while Eaton, Beck, and Pearson (1941) observed a man who during hospitalization in San Francisco transmitted psittacosis to three nurses, with two deaths. Even ward infections have been reported (Pinero Garcia, 1940). A physician, who contracted his disease from a fatal case, was visited by an intern; the intern developed psittacosis and recovered, but during his illness he infected his nurse, who died, and she in turn transmitted the virus to a second nurse.

The pathways of transmission from bird to man are threefold, and the order of importance is as follows: (a) Indirect transmission by air, (b) through handling sick or dead birds, or having contact with feathers, excreta, or nasal discharges of sick or latently infected birds, and (c) through bite wounds (Laubscher et al., 1945). The dispersion of particles of virus adherent to desiccated fecal droppings may be demonstrated readily in "sentinel experiments." By exposing ricebirds for varying lengths of time in rooms which house infected parrots, in a manner which excludes ingestion of feed contaminated with droppings, the experiences in the epidemiology of human psittacosis may be reproduced as perfect models. Contrary to general belief, actual contact with diseased parakeets or pigeons is not necessary, since air currents may disseminate particles of the virus. Birds acquire the infection, in all probability, by ingestion and occasionally by inhalation. Some experimental data suggest a germinative transfer, since yolks and eggs in the oviduct have been found to carry virus. Some of the present-day knowledge concerning psittacosis is shown graphically in Figure 22.1.

## ETIOLOGY AND PARASITOLOGY

The filtrable "virus" character of the disease agent in the splenic and hepatic emulsions from parrots and humans was established independently and in rapid succession in 1929–30 by Krumwiede and associates, and Arm-

strong and McCoy in the United States; by Levinthal in Germany; by Bedson and Western in England; and by Sacquépée in France. With filtrates which were sterile on bacteriological media, the disease was reproduced in healthy birds. Of particular importance and an invaluable aid to research on psittacosis was the discovery by Krumwiede, McGrath, and Oldenbusch that the virus is transmitted readily to white mice. Furthermore, in view of the fact that only filtrates through the coarser grades of filter candles were infectious, the virus particles were demonstrated promptly by a number of workers in the early months of 1930.

Generally described as Levinthal-Cole-Lillie bodies (L.C.L. bodies), the largest elementary bodies observed in microphotographs measure 380 mμ, and the smallest, 280 mμ (Lazarus and Meyer, 1939), while according to Kurotchkin *et al.* (1947) in electron micrographs the mean diameter of the spherical elements is 455 ± 78 mμ. The size of the feline pneumonitis virus in gold-shadowed preparations is reported as 525 mμ ± 84 (Hamre *et al.*, 1947). Moshkovsky (1945) has suggested the name *Miyagawanella psittaci* for the agent, and has grouped it in the family of Chlamydozoaceae. The microchemical reactions of the elementary bodies (readily stained by the Macchiavello or Castaneda technic for rickettsias with Giemsa, haematoxylin) and their morphology, as well as the multiplication within reticuloendothelial cells or cellular elements of tissue cultures, place the bodies more in the group of bacteria than the true viruses. By comparison with other morphologically visible viruses, their position is unique (Robinow and Bland). They are both Feulgen- and Castaneda-positive, while the virus of lymphogranuloma is only Castaneda-positive. The vaccinia particle is not demonstrable by either method.

Elementary bodies of all sizes can be seen in smears from infected pericardial or peritoneal exudates or organs of parrots, parakeets, pigeons, and ducks. Bland and Canti (1935), through painstaking microscopic studies, demonstrated that the elementary bodies undergo a developmental cycle in the cytoplasm of the host cells. Of perfect spherical shape, they resemble the Paschen bodies or certain Rickettsiae; on tissue cultures or in yolk sac cells they increase rapidly in size, and become embedded in a homogenous ground substance or matrix. These initial bodies at first divide to elements of comparable size (3 to 12μ in diameter), but as multiplication progresses the elements of division become smaller and smaller until the final elementary body stage is reached again. Yanamura and Meyer (1941) noted that within the matrix tinctorial differences may be observed; the large virus particles stain blue and the smaller forms (0.3–0.4μ) red, when stained according to the method of Castaneda. A capsular substance or thin membrane may be responsible for the varying tinctorial reactions. Usually by the 48th hour the matrix of the inclusion body presents evidences of lique-

faction, and the elementary bodies have become so numerous that they fill the entire cytoplasma of the cell. Death of the host cell and autolysis of the colony releases myriads of particulate elementary bodies, which are then capable of invading and repeating the cycle in new cells. From a historical point of view, Levinthal was the first to recognize the great variability in size of the virus particles, and proposed the name *Microbacterium multiforme psittacosis*. More recently, Moshkovsky (1945) suggested that agents of the psittacosis-lymphogranuloma group, which show an identical developmental cycle (Rake and Jones, 1942) be classified as genus Miyagawanella, family Chlamydozoaceae. This classification, somewhat misleading, has been adopted by the Board of Editor-Trustees of Bergey's *Manual of Determinative Bacteriology*, Sixth Edition (in press).

The number of Miyagawanella found in spontaneously infected birds and in man varies greatly. In all psittacosis infection, the agent invades and destroys reticulo-endothelial cells. Staining reactions and morphological appearance of the virus are so characteristic that proper identification is usually easy. It grows freely on chorio-allantoic chick membranes (Burnet and Rowntree, 1935; Lazarus and Meyer, 1939), in liquid media, or on solid media of the Zinsser-Fitzpatrick-Wei type. Although growth may be abundant, infectiousness for mice and birds declines gradually after 300 passages in tissue culture. No evidence of virus multiplication in media devoid of viable cells has been presented. Elementary bodies are deposited by fractional centrifugation in an angle machine; they are readily agglutinated by specific antisera or serve as antigens in complement-fixation tests.

Since infectivity depends on the number of elementary bodies in filtrates, and filtrations through collodion membranes which retain the elementary bodies are noninfectious, it is now generally recognized that *Miyagawanella psittaci* is the cause of psittacosis. Bedson found that the psittacosis virus contains two antigens: one a heat-labile antigen destroyed by temperatures above 60° C., the other able to withstand boiling. Upon infection, antibodies to both the heat-labile and heat-stable antigens are produced, the latter in considerably greater quantity. Although the relation of the antibodies to the heat-stable antigens in the production of immunity to the psittacosis serum is not known, these antibodies constitute a reliable index of infection. The heat-stable antigen is also ether-soluble, and in all probability is common to the protein molecule of the entire lymphogranuloma-psittacosis group (Hilleman and Nigg, 1946). Nothing definite is known about the antigens which give rise to the antibodies responsible for the highly species specific protection against different members of the lymphogranuloma-psittacosis group (Hilleman, 1945). Yolk sacs infected with the psittacosis agent, when shaken with amniotic and allantoic fluid, are toxic to mice on intravenous or intraperitoneal injection. This toxin is labile and not readily

separated from the elementary bodies. Specific antitoxins produced in rabbits and chickens are effective against a few lethal doses of the toxin.

Crude heavy suspensions in broth may remain infectious at ±4° C. for several weeks, but preparations of elementary bodies in buffered saline are noninfectious 29 days after preparation and storage. Frozen at —70° C., the virus remains active for over two years. When preserved in 50 per cent glycerol in buffered saline with a pH 7.6 and held at room temperature, heavy suspensions retain their activity for from 10 to 20 days. Sputum and human lung specimens rapidly lose potency in glycerol. Formalin 0.1 per cent and phenol 0.5 per cent inactivate the psittacosis virus in 24–36 hours, while 10 per cent ether at room temperature is destructive within 30 minutes on heavy yolk sac suspensions, or agar tissue cultures. The infectiousness of sputa or organs is markedly reduced by microbian activities.

The white mouse, universally available and relatively safe, has displaced birds for experimental purposes, except in rare instances when Java rice-birds (*Padda oryzivora*) offer advantages. Mice highly susceptible to bacteria, but not necessarily to neurotropic virus infections, are preferred. Psittacosis viruses administered by the intranasal route produce characteristic lesions in lungs of mice, guinea pigs, hamsters, cotton rats, squirrels, and monkeys. Several viruses are known to be highly pathogenic for guinea pigs by the subcutaneous and intraperitoneal route (Olson and Larson, 1945). The feline strain and certain murine strains (Grebb) excepted, the majority of psittacosis viral agents when injected intracerebrally cause irritability, motor hyperactivity, ataxia, convulsive seizures, and death within 3 to 6 days. Viruses isolated from pigeons, chickens, and ducks may be maintained through intracerebral mouse passage or in cultures. Intranasal administration in mice should be avoided, since the virus may become contaminated with pleuropneumonia organisms or psittacosis-like strains of pneumonitis virus (Gönnert, 1941; Nigg and Eaton, 1944; and DeBurgh, Jackson, and Williams, 1945).

### SPECIES OF BIRDS SPONTANEOUSLY INFECTED WITH THE PSITTACOTIC VIRUS

At least thirty-one species belonging to nineteen genera have been proved to be spontaneously infected (Table 2). Common to all is the high incidence of prolonged latent infection. Although true epizootics of the type seen occasionally in psittacine birds, in which transmissions by contact spread the infection throughout entire aviaries or cages, have not been observed, it is well known that feed from an infected pet shop may introduce psittacosis into a flock of canaries (Gerlach). Bengalese and goldfinches associated with shell parakeets suffering from latent psittacosis repeatedly contracted the

infection within two to three weeks. Experimentally, many other species have been infected successfully, but the finch varieties are aberrant hosts.

The wide distribution of psittacosis among domestic and wild pigeons (*Columba livia*) is now fully established. Approximately 30 to 40 per cent

TABLE 2

THE DISTRIBUTION OF SPONTANEOUS PSITTACOSIS INFECTION IN THE CLASS AVES

ORDER: PROCELLARIIFORMES: (1)
  FAMILY: Procellariidae: *Fulmarus glacialis* (L.), fulmar

ORDER: ANSERIFORMES: (1)
  FAMILY: Anatidae: *Anas platyrhynchos* (L.), mallard (domestic)

ORDER: CHARADIIFORMES: (2)
  FAMILY: Scolopacidae: *Catoptrophorus semipalmatus* (Gmelin), willet
  FAMILY: Laridae: *Larus argentatus smithsonianus* (Coues), American Herring gull

ORDER: COLUMBIFORMES: (3)
  FAMILY: Columbidae: *Columba livia livia* (Gmelin), tame pigeon; *Streplopelia decaocto decaocto* (Frivaldszky), ringed turtle dove; *Streplopelia semitorquata* Ruppell

ORDER: PSITTACIFORMES: (33)
  FAMILY: Psittacidae: *Trichoglossus chlorolepidotus* (Kuhl), scaly-breasted lorikeet; *Trichoglossus haematod moluccanus* (Gmelin), *Kakatoe sanguinea sanguinea* (Gould), greater blood stained cockatoo; *Kakatoe galerita galerita* (Latham), sulphur-crested cockatoo; *Kakatoe roseicapilla roseicapilla* (Vieillot), pink parrot; *Nymphicus hollandicus* (Kerr), quarrion; *Ara macao* (L.), red and yellow macaw; *Aratinga pertinax tortugensis* (Cory), parakeet; *Aratinga pertinax margaritensis* (Cory); *Nandayus nanday* (Vieillot), black-headed conure; *Forpus passerinus Spengeli* (Hartland) parrotlet; *Forpus conspicillatus conspicillatus* (Lafresnaye); *Myiopsitta monachus monachus* (Boddaert), green paroquet; *Graydidasculus brachyurus* (Kuhl); *Pionus menstruus* (L.), blue-headed parrot; *Amazona festiva festiva* (L.), festive amazon; *Amazona barbadensis barbadensis* (Gmelin); *Amazona aestiva aestiva* (L.), blue-fronted amazon; *Amazona ochrocephalus panamensis* (Cabanis), Panama yellow-headed parrot; *Amazona albifrons albifrons* (Sparrman), spectacled parrot; *Psittacus erithacus erithacus* (L.), gray parrot; *Psittacula krameri manillensis* (Beckstein), parrotlet; *Polytelis anthopeplus* (Lear), black-tailed parakeet; *Alisterus scapularis scapularis* (Lichtenstein), king parrot; *Agapornis roseicollis* (Vieillot); *Agapornis personata* (Reichenow), African love-bird; *Platycercus elegans elegans* (Gmelin), crimson parrot; *Platycercus eximius eximius* (Shaw), rosella parakeet; *Platycercus eximius cecilae* (Mathews); *Platycercus zonarius semitorquatus* (Quoy and Gainard); *Platycercus adscitus adscitus* (Latham); *Psephotus haematonotus* (Gould), grass parrot; *Melopsittacus undulatus* (Shaw), budgerigar.

ORDER: PASSERIFORMES: (14)
  FAMILY: Ploceidae: *Lagonosticta senegala* (L.), firefinch; *Munia oryzivora* (L.), Java sparrow; *Uroloncha striata; Poephila mirabilis*, (Des Murs); *Poephila gouldiae* (Gould), Gouldian finch; *Poephila acuticauda* (Gould), long-tailed finch; *Zonaeginthus guttatus* (Shaw), Diamond sparrow.
  FAMILY: Fringillidae: *Carduelis carduelis* (L.), goldfinch; *Carduelis major* (L.), goldfinch; *Spinus tristis* (L.), goldfinch; *Serinus serinus canaria* (L.), canary; *Pyrrhula pyrrhula europaea* (L.), bullfinch; *Cyanospiza ciris* (L.), painted bunting; *Parus major* (L.), great titmouse.

of nearly 1,000 samples of pigeon blood taken throughout the United States gave specific complement-fixation reactions in the presence of psittacosis antigens. Virus was readily isolated from spleens, livers, kidneys, or intestinal contents. The rate of infection in some pigeon lofts is probably higher than 40 per cent, since birds harboring the virus may give negative complement-

fixation reactions. To breeders of pigeons, fanciers of racing pigeons, and the public at large, this reservoir of disease is important. In the United States alone during the past seven years, 110 clinical infections in humans have been attributed to contact with domesticated pigeons. Conclusive proof of sub-clinical infections in persons exposed to pigeons in laboratories, recently acquired, indicates that the rate of human infections of Columbidian origin may be even higher. The role of wild pigeons in cities and towns is not known, but it is significant that rates of avian infections in large eastern cities vary between 18 and 40 per cent, while in Ontario, Canada, it is 17 per cent (Labzoffsky, 1947). Pigeons deserve to be investigated as spreaders of infection to barnyard fowl. Since the host-parasite relationship in psitta-cosis of pigeons is a symbiotic one, there is every reason to believe that Miyagawanella has for generations parasitized the pigeon, and that psittacosis is not a recently acquired infection, but only its discovery is new.

**Chickens.** The susceptibility of chickens (*Gallus gallus*) to psittacosis was anticipated in experiments by Bedson and Western, Krumwiede, *et al.,* and Levinthal, Dahmen, and Hamel, and in exposure experiments by Meyer and Eddie. A human infection traced to a chicken farm in New Jersey furnished a sample of thirty-one birds, four of which harbored viruses re-sembling pigeon strains in the organs. A dove was seen to visit the barnyard, and it was thought that the chickens contracted the infection through contact with the pigeon. Further observations have strengthened this interpretation. Epidemiologic investigation of two human cases attributed to pigeons dis-closed that the infected birds were caged over a chicken pen; two of the chickens, when examined, yielded pigeon-psittacosis viruses from organs and intestines. A small outbreak of atypical pneumonia in a children's home in Westchester, New York, attracted attention when it was learned the patients handled infected pigeons and that some had played with chickens exposed to the pigeons. The cloacal contents of three of four chickens contained pigeon viruses. Until a simple serological test for mass examinations of barnyard fowl has been developed, the extent of spontaneous psittacosis in chickens cannot be determined. The question of whether chickens acquire psittacosis through contact with pigeons or if it exists in nature as an independent infec-tion common to the species must for the present also remain unanswered.

Following recognition of human infections by Dr. William Wolins on Long Island, New York State, 115 ducks and ducklings (*Anas platyrhyn-chos*) from nine duck farms were autopsied, and pools of organs tested on mice. Thirty-eight per cent yielded psittacosis-like viruses with a patho-genicity similar to pigeon strains. Previously, psittacosis had been demon-strated in a mallard duckling which had infected the owner. Organs of three of eleven ducks obtained from a Petaluma, California, farm yielded psitta-cosis virus.

Existence of a psittacosis virus, transmissible to mice and parakeets, in newly-fledged fulmars (*Fulmar glacialis*) and petrels from the Faroe Islands was proved by Haagen and Mauer. Bedson reports that sera of five patients with a disease similar to psittacosis, sent to him from Iceland, gave positive complement-fixation reactions; fulmars are used as food there. Thus, the whole migration and breeding range of these sea birds must be considered a potential reservoir of psittacosis. Inductive epidemiological investigations of human infection have incriminated game birds, such as pheasants. The extent of psittacosis in the bird kingdom needs further investigation.

### THE SPONTANEOUS DISEASE IN BIRDS

**Psittacine birds.** Observations of the past ten years fully attest to the important fact that *the clinical manifestations in the different species of parrots, parakeets, or conures infected with psittacosis are not characteristic. In order to prove the nature of the disease, autopsy and laboratory examinations are imperative.* Equally far reaching is the epidemiologically important discovery that visibly "healthy" birds may harbor the virus and, as shedders, disseminate the infective agent, or that these chronic carriers may, when transferred to unhygienic conditions, develop fatal clinical disease. Surveys have shown that a whole spectrum of various stages of infection, from the frankly sick to the latent state, may be bridged by atypical and rudimentary cases, which present few or only temporary transitory symptoms. From an epidemiological standpoint, these cases are frequently of greater importance than the visibly sick birds. Concerning the incubation of avian psittacosis, the following data are of interest. In the injection experiments of Bedson and Western, the interval between injection and death was from 3 to 29 days. The period between exposure and death was 41, 61, and 106 days (Meyer) .

A parrot or parakeet with psittacosis is sleepy and listless and refuses to eat. Its wings droop, and its feathers stand on end, readily falling out. The bird may show fits of shivering and weakness, or may be unable to sit on the perch. This condition progresses for one or two weeks during which there is diarrhea with greenish, occasionally blood-tinged, watery droppings, and marked anorexia appears. Wasting is rapid. Death may be preceded by convulsions and paralysis. The parrot or parakeet may remain in this stage for several days or weeks, and then die suddenly or make a slow recovery. The rate of mortality among imported young parrots may be very high, while among parakeets held in captivity, it may be as low as from 2 to 5 per cent, and the cause of death may not be recognized by the breeder. As a rule, young immature birds known as "crawlers" are particularly apt to succumb.

**Gross pathological findings.** (a) *Acute stage.* There is always wasting of pectoral muscles. The skin may be covered with an erythematous rash, macules from 2 to 4 mm. in diameter being scattered uniformly over the body

and legs, 1 to 2 cm. apart. Mucous plugs are present in the nasal openings. Internally, a profuse semipurulent coating may cover the walls of the air sac, or a fibrinopurulent thickening may adhere to the inner lining of the sternum. A massive fibrinous exudate or an effusion with fibrinous flakes may cover the heart and fill the pericardial sac. Similar exudations or plastic deposits may be present over the capsule of the liver. The liver always is



FIG. 22.2 Spleens of shell parakeets removed from an aviary infected with psittacosis. Small spleens (less than 3 mm. in diameter) are noninfective for mice. Natural size.

swollen, with rounded edges, slightly pale saffron to ocher-colored, and finely mottled with shades of greenish brown or patchily discolored. Occasionally (in about 10 per cent), it is studded with fresh small and large areas of necrosis or infarction surrounded by hemorrhagic zones. The spleen as a rule is enlarged, occasionally covered with inflammatory lymph, dark in color, and sometimes spotted with fine necrosis. The kidneys are swollen, light grayish in color, and quite soft and friable. The intestinal serosa is injected;

the mucosa, hyperemic. Only in very rare instances are lesions demonstrable in the lungs, in which a few red, flabby areas of consolidation may be present.

(b) *Chronic latent stage.* Visibly healthy, well-nourished parrots or shell parakeets examined in the course of surveys may yield essentially negative autopsy findings with the exception of enlarged spleens (Fig. 22.2). Some birds with enlarged spleens may show a slight thickening of the walls of the air sac and scars of healed necrosis in the liver with peritoneal adhesions. Experience has taught that in parakeets, spleens with diameters of from 4 to 6 mm. may contain virus, while larger organs, from 7 to 12 mm. in diameter, may be noninfectious. Identical observations have been made by Burnet on grass parrots, cockatoos, cockateels, and lorikeets.

**Microscopic pathological findings.** Smears prepared from pericardial or peritoneal exudates and spleens, and stained according to the Macchiavello fuchsin-methylene blue or Castaneda methylene blue-safranin method, or with a modified Giemsa's stain, as a rule reveal numerous intracellular and free elementary bodies of *Miyagawanella psittaci* (Figs. 22.3 and 22.4). Sections of tissues, preferably fixed in Zenker's solution and stained with Giemsa's solution, haematoxylin, eosin, or Heidenhain's iron haematoxylin, make excellent preparations for the study of changes induced by the virus. In the spleen, slight alterations in the lymph follicles may be displaced in advanced stages by a complete alteration of the architecture. As a rule, the reticular and sinus structures are well preserved. Enormous infiltrations of mononuclear cells filled with amorphous debris, haemosiderin, pigment, and elementary bodies are largely responsible for the increase in size of the spleen. Vacuolation of these cells in places approached definite necrosis. Granular leukocytes usually are absent. In chronic infections, hyperplasia of the reticulo-endothelial apparatus is the predominant change which obliterates the follicles.

The characteristic necrosis in the *liver* has its onset in the death of isolated hepatic cells or groups of cells. The cytoplasm becomes oxyphilic without nuclear staining in the acini. Mononuclear phagocytes and a few polymorphonuclear cells surround and infiltrate these areas. With the disintegration of the necrotic cells of the liver, strands of acidophilic, hyaline material, collections of leukocytes and depositions of fibrin are left in the supporting tissues. Extensions of these areas lead to accumulations of inflammatory cells under the Glisson's capsule and to a perihepatitis. Proliferation of the cells of the liver is recognized readily by mitotic figures. Throughout the liver, hyperplasia of the Kupffer's cells and focal accumulations of wandering mononuclear cells with vacuolated cytoplasma contain varying numbers of Miyagawanella. Plasma cells are numerous. The bile ducts are frequently dilated, the epithelia desquamated and filled with mononuclear phagocytes. As a rule, the vascular apparatus is not markedly damaged,

although fibrinous thrombi are seen in the small vessels; necrosis of the hepatic cells hardly can be attributed to these thrombi. In chronic and in healing lesions, irregularly shaped collections of hepatic cells are separated by proliferating bile ducts, foci of lymphocytes, and fibrous tissue.

In the *kidneys*, the epithelium of the secretory tubes and glomerular capsules may be packed with virus particles. With the destruction of the tubular epithelium, a margin of actively proliferating, stellate epithelioid cells, lymphocytes, and a few eosinophilic leukocytes make their appearance. Around these granulomatoid areas and among the tubules, a dense



FIG. 22.3. Intracellular colonies of *Miyagawanella psittaci* in pericardial exudate of a pigeon. Approximately ×1,200.

interstitial infiltration with lymphoid cells, plasma cells, and a few leukocytes creates readily recognizable focal lesions, which doubtless contaminate the urinary secretion with the infective agent. Occasionally the ureters show lymphocytic and plasma cell infiltrations in the mucosa, and partial desquamation of the lining epithelium. The cytoplasma of these cells are filled completely with coccoid and bacillary elementary bodies. In the *lungs*, intracellular vacuolation and edema may be associated with patchy serous exudation in the atrial air cells; the scattered lobular inflammation and consolidation of the parenchyma seen in man are exceptionally rare. The lumen of the intestines is filled with desquamated epithelial cells containing elementary bodies. Lymphoid cells infiltrate the villi in which the columnar epithelia are the host cells for inclusions. Acute psittacosis of parrots and, to a lesser degree, also of parakeets is characterized by a desquamative catarrhal enteritis; a desquamative and interstitial ureteritis; a focal tubular, granulomatous

nephritis; a desquamative and ulcerative cholangitis with focal necroses grading into granulomata in the liver, with associated interstitial lymphoid infiltration; reticulo-endothelial swelling, hyperplasia, and phagocytic activity in the liver, spleen, and bone marrow; and occasional focal proliferative, desquamative, and cellular exudative inflammation of the various serous membranes (Lillie) .

**Finches and canaries.** Ricebirds, canaries, and various other species of the order PASSERIFORMES contract psittacosis when exposed in cages or rooms where infected parrots or parakeets are kept. After a variable incubation time
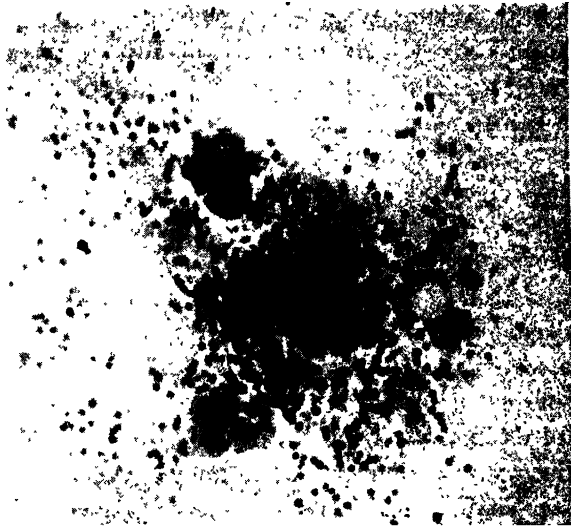


FIG. 22.4. *Miyagawanella psittaci*—elementary forms set free from a crushed cell—peritoneal exudate of a mouse. Approximately ×1,400.

of from 15 to 60 days and a period of from 3 to 20 days, the birds appear listless and huddle, with ruffled plumage, in the corner of the cage. Weakness, anorexia, and emaciation may not be noticed on account of the acute and sudden death which terminates the infection. At autopsy, the anus invariably is soiled with greenish or grayish fecal concretions, and the subcutaneous tissues and serous linings are jaundiced. The liver is enlarged, fatty, irregularly mottled yellow, and in a few instances studded with small necroses. The spleen is always enlarged, soft, and sausage-like, and thickened (from 15 to 18 mm. long and from 4 to 8 mm. thick) ; the kidneys are swollen, grayish, and soft; urine accumulates in the cloaca; the serous lining of the sternum, air sacs, pericardium, and liver are frequently covered by a plastic purulent exudate. The intestines are injected and in part obstructed by liquid chyme. In very protracted infections, focal pneumonic areas have been encountered, and a general hyperemia of the trachea accompanies this process. The micro-

scopic pathologic changes are quite similar to those described for psittacine birds.

**Pigeons.** J. D. W. A. Coles, at Onderstepoort, South Africa, described a psittacosis of pigeons associated with *Haemoproteus columbae, Trichomonas hepatica* and *Salmonella typhimurium.* The birds were visibly sick, listless, and showed no interest in food; vent feathers were soiled with liquid feces. At autopsy the lungs were edematous, the liver and spleen swollen. There was moderate aerocystitis in the abdominal air sac, and moderate catarrh of the intestines. Elementary bodies were demonstrated in the blood, lungs, and spleen, and the virus was obtained through mouse passage. Andrewes and Mills (1943) isolated several strains from pigeons in Southern England.

In the United States, acute spontaneous infections have been observed by Pinkerton and Swank, and by Meyer, Eddie, and Yanamura in pigeons fed thiamine-deficient diets, or housed unhygienically; in reality, these infections were relapses. An outbreak of psittacosis observed by Smadel, Wall, and Gregg (1943), and an epizootic in a pigeon platoon, investigated by Smadel, Jackson, and Harman (1945) disclosed a new virus immunologically different from psittacosis, which produces intranuclear, herpes-like inclusions; and studies of pigeon lofts reporting death from psittacosis disclosed concurrent Salmonella infections. Consequently, it is frequently impossible to assess the etiologic importance of one infective agent or another as the cause of death. Observations by Meyer and associates indicate that the majority of birds in any pigeon loft acquire early infections. Some birds are visibly sick but recover, establishing latency of the virus. Infections in squabs may be entirely subclinical; in fact, *no complement-fixing antibodies may be demonstrated* despite distinct anatomical lesions, or heavily infected organs. Such pigeons are highly infective to man.

So far, only a few acute spontaneous psittacosis infections, uncomplicated with salmonellosis, have been available for examination. The symptoms are by no means characteristic. Aside from listlessness, weakness, and occasional opisthotonos, lack of appetite from several days to one week, and diarrhea, no signs which would aid in making a differential diagnosis are apparent. At autopsy, plastic exudates over and inside the pericardium and over the liver are usually present. The livers are swollen and hemorrhagic, occasionally mottled, while the moderately enlarged spleen is dark pink. Parenchymatous changes are always present in the kidneys. A catarrhal enteritis of varying degree with considerable accumulation of urates in the cloaca likewise may be noted. In the few pigeons with caseous pneumonic foci, Salmonella organisms were isolated invariably from lesions. Pulmonary lesions are in all probability not a part of the psittacotic infection. Smears prepared from the fibrinopurulent exudate present in the pericardium or over the liver are usually rich in virus particles, either free or in the cyto-

plasma of monocytic cells. Organs and exudates derived from pure psitta-
cosis of pigeons are sterile when planted in ordinary media or enriched in
brilliant green broth.

The majority of psittacosis infections in pigeons are chronic and sub-
clinical. Complement-fixation tests reveal that flocks responsible for human
infections are invariably very heavily infected; for example, fifty in a lot of
sixty, and sixteen of a lot of eighteen, gave strongly positive reactions. In
sacrificed birds, chronic infection may be recognized by enlarged, dark
purplish, mottled, or pale spleens (Fig. 22.5), and soft, slightly grayish kid-



FIG. 22.5. A—spleens of pigeons proved to be infected with psittacosis. Natural size.
B—two spleens and one liver of mice infected with psittacosis virus; necrosis in liver. Small
spleen not infected. Natural size.

neys, with focal hyperplasia of the reticulo-endothelial elements or peri-
tubular lymphoid infiltration.

In the subacute stages, the residual evidence of a pericarditis may still be
present, and its psittacotic origin may be proved by intracerebral injection of
a suspension of the exudate into mice. With some difficulty, the presence of
the virus in the spleen or kidneys may be established through intranasal or
intracerebral mouse inoculation of organ suspensions. Pigeons with or with-
out complement-fixing antibodies in their sera may yield the virus. With a
very few exceptions, pigeons from commercial pigeon lofts are immune to
intramuscular infection with pigeon psittacosis viruses, but they develop a
fatal meningo-encephalitis when injected intracerebrally. The remarkable re-
sistance of this species of bird to psittacotic virus by feeding or intramuscular
injection is attributable in all probability to an acquired immunity fre-
quently conditioned by the persistence of the infective agent in the tissues.

Doves (*Streplopelia risoria*) as a rule are free from psittacosis, but re-
cently birds bought in the open market or associated with parakeets and other
birds in aviaries were proved to be infected, by complement-fixation tests
and isolation of the virus. Several species have acquired the disease through
association with psittacine birds in zoological gardens (Tomlinson, 1941).

Spontaneous psittacosis infection of *chickens* was discovered by Meyer and Eddie on a poultry farm in New Jersey in 1940 in connection with a human infection; for some time the owner had been losing approximately 500 chickens of the hatchings from "range paralysis." The patient, furthermore, had dressed the chickens and had noted that cadavers were emaciated and often had large livers. In a sample of twelve chickens, two gave specific agglutination tests with formalinized psittacotic antigens, and it was deemed advisable, therefore, to test the organs of twelve birds for virus on mice. The autopsies of thirty-one chickens with few exceptions revealed no gross lesions suggestive of psittacosis, although anemia, emaciation, and slightly enlarged spleens were noted in five. Through inoculation tests with organ emulsions which were sterile on lifeless media, a virus indistinguishable from that obtained from pigeons was isolated from four. Psittacosis virus has been isolated from the organs and intestinal contents of chickens in contact with pigeons, although anatomical lesions suggestive of psittacosis were not found. Without tedious inoculation tests on mice, the shedder and carrier stages of Miyagawanella in chickens would not have been discovered. That chickens closely associated with infected pigeons or parakeets may acquire the disease was suggested in an exposure experiment with ten Dominick Plymouth chickens (four to six months old) reported by Meyer (1935). Those held in pens with sick or latently infected parakeets (40 per cent) died on the fifty-second and sixty-third days of the exposure period, and there were four latent infections with typical lesions.

**Pekin ducks.** Although psittacosis virus has been demonstrated in organs and intestinal contents of Pekin ducks, the importance of the findings to the duck raising industry has not been assessed as yet. There is definite evidence that the virus, present in approximately one-third of the bird population, occasionally infects workers on commercial farms, or persons who keep ducks as pets. The data thus far collected are incomplete, and in no way suggest a hasty conclusion that psittacosis is responsible for serious bird mortalities which sporadically plague the duck industry. There are facts supporting this statement; in a series of six dead ducklings, 4 days old, the agent was isolated from the spleen, liver, and kidney of only two. In another group of fifty-three sick ducks of various ages, it was demonstrated in twenty, or 37.7 per cent, and was found only in the intestines of five, or 9 per cent. Of thirty-three healthy ducks serving as controls, seven, or 21 per cent, harbored virus in the intestines and four, or 12 per cent, in the organs. Differences between sick and healthy ducks were not significant enough to warrant the conclusion that sickness or death was caused by Miyagawanella agents. Extensive investigations, including bacteriological and supplementary virus examinations, must be made before the importance of psittacosis in the pathogenesis of duck diseases is established; thus far, there is no

evidence that it causes epizootics. Ducks are susceptible to Salmonella and other infections. Organs and intestines of twenty-three of ninety-seven ducks cultured before being tested for virus yielded *Salmonella typhimurium; Pasteurella avicida* and *Pfeifferella anatipestifer* (Hendrickson and Hilbert, 1932) were found in two others. Anatomical lesions were bacteriologically sterile in six specimens and infected with Salmonella in four others which subsequently produced psittacosis in mice. As in psittacosis of pigeons, clinically normal ducks revealed no gross anatomical lesions despite presence of virus in the organs. That the agents were found in cloacal contents of ducks whose organs proved singularly free from virus attests to the existence of shedders, and suggests that psittacotic parasites are constantly being ingested from an environment commonly contaminated with excreta. Demonstration of the agent in the organs of a 4-day-old duckling dead of primary salmonellosis offers proof of liberal exchange of infectious agents discharged in the feces. The available facts about psittacosis on commercial duck farms tentatively suggest that there is intimate parasitism between a psittacosis virus of low virulence and the bird population. The agent may cause occasional deaths, and may reduce resistance to bacterial infections and possibly other viruses frequently found in such establishments.

**Fulmars.** Nothing definite is known concerning psittacosis infections in fulmars. The report by Haagen and Mauer on the isolation of virus gave no description of anatomical lesions. Some birds were doubtless virus carriers, but whether or not any become visibly diseased is still unknown. Only young fulmars have been found to be infected.

**Laboratory diagnosis.** Since it is impossible to diagnose psittacosis clinically or at autopsy, the public health bacteriologist or clinical pathologist must study the laboratory diagnosis of the disease (Fig. 22.6). Briefly, the technique is as follows: Any material containing psittacosis virus must be regarded as highly pathogenic and dangerous to handle unless proper precautions are taken. Birds scatter infected material in dried feces and nasal discharges attached to feathers and particles of down. Dead birds or birds killed with chloroform should be immersed completely with the cage into 5 per cent lysol solution, wrapped in lysol-soaked cheesecloth, frozen with dry ice, and preferably sent to a specially equipped laboratory. If the pathologist wishes to conduct his own examinations, autopsies should be conducted in a special room where there is no chance of the infective material becoming dried. A special gown, rubber gloves, and a suitable face mask with goggles should be worn. Inoculated mice should be kept in glass jars, with perforated metal lids, which are covered with several layers of gauze to prevent escape of dust. Experiments on birds should not be undertaken by inexperienced pathologists.

Smears prepared during autopsy from pericardium, serous surface of the

# STEPS TO DETERMINE THE PRESENCE
## OF PSITTACOSIS VIRAL AGENTS
## INFECTED BIRDS (OR MAMMALS)



Heart
Lung
Liver
Gizzard
Spleen

Lung
Liver
Gizzard
Enlarged spleen

Pericardial exudate

Liver necrosis

Enlarged spleen

Exudate on intestines

Cloacal content

*NORMAL*                                 *INFECTED*

Smears stained for elementary bodies

Cloacal content

exudate spleen liver kidney

Emulsify and dilute
with broth or anti-
biotic in grinder
15-10 per cent

Culture on blood
agar and enteric
media

*Refrigerator*
overnight

If heavily contaminated *centrifuge* or filter

Inject

Death    Sacrifice

5 mice

*Autopsy* see under human specimens

Fig. 22.6. Graphic presentation of the technical procedures employed in the diagnosis of psittacosis in birds.

liver, hepatic necroses, or spleen are fixed for 5 minutes with methyl alcohol and then stained with a reliable brand of Giemsa stain for from 3 to 20 hours (one drop to 5 cc. of absolutely neutral distilled water). The slides may be differentiated in absolute alcohol, rinsed in water, dried, and examined. For rapid examination, the staining methods of Castaneda or of Macchiavello for rickettsia are very useful. Particularly the latter method gives excellent preparations when used as follows: A 0.25 per cent solution of basic fuchsin in distilled water with a pH of 7.4 is prepared by the addition of sodium carbonate. The tissue smear is dried gently by heat, and the fuchsin solution is filtered over the preparation through a coarse filter paper in a funnel. The fuchsin is left on the slide for 4 minutes, and is washed off very rapidly by dipping the slide in a solution of 0.5 per cent citric acid held in a Coplin jar. The acid solution is washed off very rapidly with tap water. The smear is then stained for about 10 seconds with 1.0 per cent aqueous solution of methylene blue. With a little practice, the method gives an excellent contrast stain, the Miyagawanella intracellular and extracellular being stained mostly red, while the cellular elements are blue.

If microscopical examination is negative as, for example, in latent infections, pieces of spleen, liver, and kidney are ground up with carborundum or sand to a paste, and diluted with broth to a rather thick suspension. Cultures in liquid media and on blood plates must be made in order to detect bacterial infection (salmonellosis, streptococcicosis, etc.). Then 0.5 cc. of the suspension is injected intraperitoneally into each of at least four mice. Since psittacosis viruses from pigeons are of low pathogenicity for mice when inoculated intraperitoneally, it is frequently necessary to establish infection by intracerebral inoculation of 0.03 ml. of the sterile suspension. Contaminated suspensions should then be passed through collodion membranes with an average pore size of from 450 to 600 mμ. The inoculated mice should be kept under observation for 20 days, then killed, and emulsions of their spleens inoculated into a fresh pair of mice, either by the intraperitoneal, nasal, or intracerebral route. A combination of the intracerebral and intraperitoneal modes of infection is particularly valuable. The blood of patients may contain the virus during the first 4 days of illness; during relapses it may be tested in a defibrinated state by intraperitoneal injection in quantities of 0.5 to 1.0 cc. The patient's sputum in early and sometimes in late stages of the disease is most likely to give positive results. An emulsion in broth should be extracted by repeated shaking and standing in the refrigerator for 12 to 24 hours. By injecting at least four mice, one or two may escape the concomitant infection with pneumococci and survive long enough for the development of the virus. Sometimes filtrates of the emulsion must be made in order to remove bacteria; it is then advisable to inject the mice repeatedly

on 3 successive days with not less than 1.0 cc. This procedure gives some chance to detect minute amounts of psittacosis virus. Specimens of sputum containing pneumococci may in the unfiltered state be tested on ricebirds (*Munia orizivora*) or treated with an antibiotic, bacteriostatic agent according to the method of Morgan and Wiseman (1946).

The experimental disease in the mouse is not very characteristic; symptoms consist of ruffled fur, closed eyes with discharge, apathy, and occasionally diarrhea. The duration depends on the amount of virus, its infectiousness, and the strain of mice. If death occurs within 5 to 10 days, the abdominal cavity shows a perihepatic exudate rich in virus particles. The liver may show necrosis, and the spleen is enlarged (Fig. 22.5). If death occurs between 15 and 30 days, the abdominal cavity contains a turbid serous effusion; the liver and spleen may be enlarged; virus bodies usually are scanty. Mice inoculated intraperitoneally usually show merely an enlarged spleen. Emulsions of these organs, in which the Miyagawanella is rarely demonstrable, produce on intracerebral injection a choriomeningitis clinically recognizable by the paralysis it induces. Smears prepared from the choroid plexus usually show an enormous number of virus particles. Material from the brain usually gives cultures in tissue media of the Li-Rivers type. It grows well in the yolk-sac epithelia, or produces on intranasal administration extensive lobular focal pneumonias in mice.

**Field specimens.** Field specimens or mouse material under test for psittacosis are held in Lusteriod tubes at −70° C. A viral agent whose tinctorial characteristics and developmental cycle are similar in smears prepared from the infected mouse, bird, or chick embryo yolk sac may be readily confirmed as belonging to the psittacosis group by complement-fixation test. Serum neutralization tests according to the method of Hilleman (1945) and St. John and Gordon (1947), with antisera produced on chickens by intensive hyperimmunization, conclusively prove that psittacosis viruses are identical with the meningopneumonitis virus isolated by Francis and Magill. Sera prepared against a mouse pneumonitis virus neutralized two mouse viruses, but had no effect on a human or parakeet-psittacosis agent. According to Rake and Jones (1944), and Hamre and Rake (1944) the endotoxins in yolk-sac suspensions of psittacosis viruses are specifically neutralized only by homologous antiserum.

**Complement-fixation tests.** The complement-fixation test developed by Bedson and modified by Meyer, Eddie, and Yanamura, and Smadel, Wertman, and Reagan (1943) has aided in the early diagnosis of psittacosis. Cocto-killed antigens prepared from infected mouse spleens may be used, but ether-killed purified tissue cultures grown on Zinsser-Fitzpatrick-Wei media, or infected yolk sacs from embryonated hen's eggs are preferred. With

true psittacosis, there is a reaction of 1:4 on the eighth day and invariably a rapidly rising titer up to 1:256 within the next 15 days. In many patients, persistence of antibodies has permitted confirmation of the clinical diagnosis in retrospect. Infected psittacine birds, finches, and pigeons give definite complement-fixation reactions. They may be strongly (1:128++++) to moderately (1:2 to 1:4++++) positive in birds which harbor the virus in a latent state in the spleen, liver, and kidney. Immature parakeets, however, and a variable percentage of pigeons with virus demonstrable in the tissues may not contain antibodies. Irrespective of these apparent discrepancies, the complement-fixation test is most useful in the detection of psittacosis in an aviary, or in a pigeon loft. Mouse inoculation tests have proved that birds in a flock.yielding from 10 to 20 per cent positive sera invariably are infected.

The use of the complement-fixation test in the epizootiology of avian psittacosis has two disadvantages: (a) The handling and bleeding of parrots endangers those who are not immune and should be entrusted only to persons who give serological evidence of a passed inapparent infection, or to those who wear properly constructed masks. (b) It is impractical to secure enough blood from the wing veins of birds the size of parakeets; but since the legal control measures require laboratory examination of the sacrificed birds, the blood tests supplement anatomical inspections and inoculation tests on mice. To protect valuable bird collections in zoological gardens, the routine quarantine procedures to which imported birds customarily are subjected may be shortened, and may be made more effective through application of serum tests. They are invaluable in establishing the existence of infection in pigeon lofts. The technique for testing chicken and duck sera has not as yet been perfected; in its place the agglutination test gives useful information.

## PSITTACOSIS IN MAN

It is difficult to be certain of the exact time of infections, but based on epidemiological studies it appears to be most often from 7 to 15 days. Since incubationary latency cannot be excluded, it is not surprising to observe occasionally an incubation time of from 30 to 40 days. The first symptoms of the disease are often vague, consisting of headache, backache, malaise, vague pains in the limbs, abdominal distention, anorexia, nausea and vomiting, thirst, excessive sweating, and photophobia. In some cases, the onset is gradual and insidious. Simultaneously with the rise of temperature, general aches and pains are likely to localize in the lumbar regions, and intense throbbing frontal or occipital headaches make their appearance. Towards the end of the first week, the whole aspect of the disease becomes more severe, and signs in the form of patchy migrating areas of consolidation, involving at

first one segment of one, and then the major part of one or both lobes of the lung, with little involvement of the bronchi and sputum, are constant features. Obstinate constipation, profound exhaustion, somnolence, and toxemia entirely out of proportion to the clinical findings predominate. At the end of two to three weeks, the temperature begins to fall by lysis and the patient gradually improves, but convalescence is protracted, tedious, and may be interrupted by relapses. The temperature rises to the level of that in typhoid fever and, with the exception of mild, abortive infections, maintains itself at a high level with only slight morning remissions. Towards the end of the second or third week, it may fall by lysis, but only in exceptional instances, by crisis. Except in the ambulatory cases, pulmonary involvement is an essential feature, manifesting itself by objective, even slight signs, and not by symptoms. A few crepitations may be heard and percussion is impaired slightly. Roentgenologic examination shows a creeping, wandering type of patchy pneumonia or pneumonitis. One of the most remarkable features is the frequent absence of rapid or deep breathing, even when physical signs in the lung indicate progressive and wide focal involvement. A slight, irritating cough may increase to painful paroxysms. Even in the presence of extensive lung signs, cough may be absent. Sputum is very scanty, entirely out of proportion to what the signs in the lungs would lead one to expect. This suggested the designation of *sputumless or nonproductive pneumonia*. Another characteristic feature of the disease is the relative slowness of the pulse except in fatal cases, in which circulatory collapse, with rapid and feeble pulse, is quite common. Constipation may continue throughout the illness, but occasionally patients suffer from diarrhea. The spleen is not palpable; the liver may descend one or two fingerbreadths below the costal margin. The absence of leukocytosis is noteworthy, and during the height of the disease a distinct shift of the neutrophils to the left is accompanied by a distinct fall in lymphocytes. Convalescence in severe cases is slow and frequently interrupted by relapses. Thrombosis of the femoral veins and postinfectious myocarditis have been seen. Definite as is the clinical entity of psittacosis in man, its certain diagnosis may, nevertheless, in any one single case and in any one stage of such a case, offer considerable difficulties. Influenza is unquestionably the disease most difficult to differentiate. Roentgenologic observation of the lung may help, and whenever there is a history of association with birds it is well to be biased in favor of psittacosis. The complement-fixation test of the patient's serum furnishes strongly suggestive evidence early in the course of the infection. Serum in an active phase will give a strongly positive reaction in a dilution of 1:8 on the tenth day, which will rise rapidly to 1:64 and higher on the fifteenth day of the disease. Inoculation of mice with specimens of sputum may decide the ultimate diagnosis, but in retrospect.

At autopsy, there is a general septicemia with an inflammatory condition of the lungs. Although grossly lobar, the uncomplicated process is lobular in distribution and not markedly related to the bronchioles. All stages of congestion, edema, and hepatization with different exudates, largely consisting of monocytes and epithelial cells, fill the vesicles. Interstitial infiltration caused by lymphocytes and monocytes and necrosis of the septa may be seen. Epithelial cells and monocytes carry the *Miyagawanella psittaci*. Pleural reactions are rare. There are usually signs of inflammation in the pharynx, larynx, and trachea, particularly in cases complicated by secondary bacterial infection. The enlarged and soft spleen shows congestion and proliferation of the fixed and free phagocytes. Parenchymatous degeneration is noted in the liver, kidneys, and myocardium, and focal necrosis and hyperplasia of the endothelia are present. Lesions suggestive of cerebral purpura often are present in the brain.

Successful treatment of human psittacosis with penicillin has been reported, and is generally considered very effective. The dynamic action of the antibiotic on psittacosis specifies a high dose level (40,000 to 100,000 units every 4 hours) in rapid succession, early in the course of the infection, in order to reduce the number of viral elements and enable the normal body defense mechanism to dispose of the few stragglers. Adherence to these principles will greatly reduce the unduly high fatality rate from psittacosis, and prevent development of virus carriers.

## IMMUNITY AND ACTIVE IMMUNIZATION

The phenomena of immunity in psittacosis are not clearly understood. It is known, of course, that parrots are actively immune following an attack. This immunity is frequently of the nonsterile type, and it is believed that the persistence of the virus is obligatory for the immunity. More recent studies, however, indicate that latency is the sequel of an incomplete or delayed mechanism of autosterilization. In fact, considerable evidence supports the belief that the resistance of the birds is in all probability innately constitutional. Individual birds may resist massive infection of virus, readily destroy it, and never become carriers, while others either succumb to small doses or recover, only to continue to harbor the infective agent in their organs for years. Since constitutional factors determine the ability of birds to resist infection, it should be possible through selective breeding to develop strains of parakeets and pigeons which will not acquire a nonsterile immunity, so eminently a source of the persistence of infection in aviaries and lofts.

Formalin-treated concentrated suspensions of psittacosis virus confer a considerable degree of immunity to parakeets and ricebirds (Meyer, Eddie, and Yanamura, 1942). This active immunity converts an apparent, often

fatal infection into an inapparent one. The facts thus far available, however, fail to encourage the hope for a practical and safe method of active immunization of birds against psittacosis.

## PROTECTIVE MEASURES AGAINST PSITTACOSIS

Psittacosis has advanced to the position of an important public health problem which could be controlled readily, provided the public would appreciate the possible danger from contact with birds of unknown origin. Importations of tropical psittacine birds may be regulated by strictly enforced quarantine measures. The period of isolation should be at least six months. The United States Public Health Service enforces such a quarantine for all importations of psittacine birds. This period of segregation may in time be shortened in the case of large parrots which can be bled readily from the veins; their sera may be subjected to the complement-fixation test. Birds which give positive reactions must either be destroyed or held until their sera become negative; nonreacting parrots must be retested within two months. If no antibodies develop in their sera, they are not infected and may be released safely. Experience has taught that, despite a properly executed health certificate without a laboratory test, untested parrots may as avian carriers introduce the infection into noninfected aviaries or dealer's stock. Exclusion of psittacosis from shipments of parakeets, parrotlets, and conures, or eradication of endemic infections from breeding establishments, though a thankless task, is within the power of a health department. The administrative and laboratory procedures are slightly different. The California plan, which has as its ultimate aim complete eradication of the avian disease from aviaries and distribution and sale of healthy pets on the North American continent, has been tried for the past six years with increasing success. In principle, it has operated as follows: Any person or party wishing to sell or barter parakeets must (a) obtain a license and receive a certificate of registration, and (b) must submit a sample of 10 to 20 per cent of his breeding stock and immature young birds to laboratory examination. Aviaries yielding infected birds are placed promptly in quarantine, and steps are taken to destroy the entire flock and to disinfect the premises thoroughly. The extent of latent infections in aviaries and progress of the sanitation program is well illustrated by the data in Table 3, which cover post-mortem examinations of approximately 26,000 parakeets.

Experience has taught that aviaries which harbor infected budgerigars may have a very low mortality from actual psittacosis. This is particularly true when the sexes are held in separate pens, and breeding operations have ceased. With the resumption of these activities, however, psittacosis reappears in a few of the immature birds. Thus, the contagium tenaciously

persists for years, and in time as many as 40 to 80 per cent of the young para-keets may show residuals of latent infections. Recent observations indicate that one test, based on a 20 per cent sample, will not always detect the exist-ence of psittacosis in an aviary. Annual retests, however, and more rigid supervision of dishonest breeders who, unknown to the control authority, introduce parakeets from private untested aviaries, should in time eradicate infected stocks.

During the war, when control measures were inadequately enforced,

TABLE 3

EXAMINATION OF CALIFORNIA AVIARIES

| | 1932 | 1933 | 1934 | 1935 | 1936 | 1937 | 1938 | 1939 | 1940 | 1941 |
|---|---|---|---|---|---|---|---|---|---|---|
| Aviaries with infected parakeets ..... | 27 (100%) | 23 (44.2%) | 47 (23.9%) | 4 (3.7%) | 1 (2.7%) | 1 (3.8%) | 3 (16.6%) | ...... .... | 1 (7.6%) | 8 (6.3%) |
| Aviaries with noninfected parakeets ..... | ..... | 29 | 136 | 109 | 38 | 26 | 18 | 13 | 12 | 126 |
| Aviaries with anatomically suspicious parakeets ..... | ...... | ....... | 12 | ...... | 8 | 6 | ...... | ...... | ..... | 1 |
| Aviaries with inconclusive findings...... | ...... | ... | 1 | 18 | 5 | ...... | ....... | ...... | ...... | 3 |
| Total aviaries tested.... | 27 | 52 | 196 | 131 | 52 | 33 | 21 | 13 | 13 | 138 |

psittacosis in breeding establishments throughout the country correspond-ingly increased. Consequently, the United States Public Health Service enacted on June 15, 1947, new regulations (Section 12.22) which limit inter-state shipments to two psittacine birds, provided they are accompanied by the owner, have been in his possession for the preceding two years, have not had contact with other psittacine birds during that period, will be transported immediately to the owner's private residence and retained there as household pets, and are accompanied by a permit from the state health department of the state of destination.

Plans to control psittacosis in pigeon-breeding establishments and back-yard lofts have not yet been evolved. In California, whenever complement-fixation tests show that the rate of infection exceeds 10 per cent, destruction of flocks is enforced.

REFERENCES

Andrewes, C. H., and Mills, K. C.: 1913. Psittacosis (ornithosis) virus in English pigeons. Lancet 1:292.

Armstrong, C., McCoy, G. W., and Branham, S. E.: 1930. Filtrability of the infective agent of psittacosis in birds. Pub. Health Rep. 45:725.

Aujaleu, E., and Jude, A.: 1936. Une petite épidémie localisée de psittacose. Presse méd. 44:1094.

Barros, E.: 1940. La Psitacose Durante el Decenio 1929–1939. Tipografico de A. Guide Buffarini, Buenos Aires.

Bedson, S. P.: 1936. Observations bearing on the antigenic composition of psittacosis virus. Brit. Jour. Exper. Path. 17:109.

————: 1940. Virus disease acquired from animals. Lancet 2:577.

———— and Western, G. T.: 1930. Psittacosis. Brit. Med. Jour. 1:882.

Bland, J. O. W., and Canti, R. G.: 1935. The growth and development of psittacosis virus in tissue cultures. Jour. Path. and Bact. 40:231.

Burnet, F. M.: 1935. Enzootic psittacosis amongst wild Australian parrots. Jour. Hyg. 35:412.

————: 1939. Note on occurrence of fatal psittacosis in parrots living in wild state. Med. Jour. Australia 1:545.

———— and MacNamara, J.: 1936. Human psittacosis in Australia. Med. Jour. Australia 2:84.

———— and Rountree, P. M.: 1935. Psittacosis in the developing egg. Jour. Path. and Bact. 40:471.

Coles, J. D. W. A.: 1941. Psittacosis in domestic pigeons. Onderstepoort Jour. (Union of South Africa) 15:141.

De Burgh, P., Jackson, A. V., and Williams, S. E.: 1945. Spontaneous infection of laboratory mice with a psittacosis-like organism. Australian Jour. Exper. Biol. and Med. Sci. 23:107.

Eaton, M. D., Beck, M. D., and Pearson, H. E.: 1941. A virus from cases of atypical pneumonia; relation to the viruses of meningopneumonitis and psittacosis. Jour. Exper. Med. 73:641.

Eddie, B., and Meyer, K. F.: 1946. Unpublished data.

Elkeles, G., and Barros, E.: 1931. Die Psittacosis (Papageienkrankheit) mit besonderer Berücksichtigung in der Pandemie des Jahres 1929/1930. Ergebn. d. Hyg., Bakt., Immunitätsforsch. u. exper. Therap. 12:529.

Fortner, J., and Pfaffenberg, R.: 1934. Über das gehäufte Wiederauftreten der Psittakose. Zeitschr. f. Hyg. u. Infektionskr. 116:397.

———— and Pfaffenberg, R.: 1935. Über das gehäufte Wiederauftreten der Psittakose. II. Mitteilung. Zeitschr. f. Hyg. u. Infektionskr. 117:286.

Francis, Jr., T., and Magill, T. P.: 1938. An unidentified virus producing acute meningitis and pneumonitis in experimental animals. Jour. Exper. Med. 68:147.

Gerlach, F.: 1936a. Beobachtungen bei der in Österreich auftretenden Psittakose. Zeitschr. f. Hyg. u. Infektionskr. 118:574.

————: 1936b. Menschen als Psittakosevirusträger nach "stummer" Infektion mit Psittakosevirus. Zeitschr. f. Hyg. u. Infektionskr. 118:709.

Gönnert, R.: 1941. Die Bronchopneumonie, eineneue Viruskrankheit der Maus. Zentralbl. f. Bakt., Abt. I., Orig. 147:161.

Haagen, E., and Krückeberg, B.: 1937. Zum Psittakoseproblem. Betrachtungen auf Grund von Beobachtungen und Untersuchungen im Jahre 1935/1936. Veröffentl. a. d. Geb. d. Volksgesundhdienstes. 48:381.

———— and Mauer, G.: 1938. Ueber eine auf den Menschen übertragbare Viruskrankheit bei Sturmvögeln und ihre Beziehung zur Psittakose. Zentralbl. f. Bakt., Abt. I., Orig. 143:81.

———— and Mauer, G.: 1939. Die Psittakose in Deutschland. (Bericht für das Altreich auf Grund der Untersuchungen im Institut "Robert Koch" für das Jahre 1937/1938.) Deutsch. med. Wochenschr. 65:808.

Hamel, C.: 1932. Quelques cas recents de psittacose en Allemagne. Bul. Office internat. d'hyg. pub., 24:966.

Hamre, D. M., and Rake, G.: 1944. Feline pneumonitis (Baker), a new member of the lymphogranuloma-psittacosis group of agents. Jour. Infect. Dis. 74:206.

————, Rake, H., and Rake, G.: 1947. Morphological and other characteristics of the agent of feline pneumonitis grown in the allantoic cavity of the chick embryo. Jour. Exper. Med. 86:1.

Hendrickson, J. M., and Hilbert, K. F.: 1932. A new and serious septicemic disease of young ducks with a description of the causative organism Pfeifferella anatispestifer, N. Sp. Cornell Vet. 22:239.

Hilleman, M. R.: 1945. Immunological studies on the psittacosis-lymphogranuloma group of viral agents. Jour. Infect. Dis. 76:96.

————— and Gordon, F. B.: 1944. Immunologic relations of psittacosis-lymphogranuloma group of viral agents. Proc. Soc. Exper. Biol. and Med. 56:159.

————— and Nigg, C.: 1946. Studies on lymphogranuloma venerum complement fixing antigens. III. The solubility in ether of an active fraction. Jour. Immunol. 53:201.

Hoge, V. M.: 1934. Psittacosis in the United States; incidence, scientific aspects, and administrative control measures. Pub. Health Rep. 49:451.

Krumwiede, C.: 1930. The virus of psittacosis. Jour. Am. Med. Assn. 94:995.

—————, McGrath, M., and Oldenbusch, C.: 1930. The etiology of the disease psittacosis. Science 71:262.

Kurotchkin, T. J., Libby, R. L., Gagnon, E., and Cox, H. R.: 1947. Size and morphology of the elementary bodies of the psittacosis-lymphogranuloma group of viruses. Jour. Immunol. 55:283.

Labzoffsky, A.: 1947. Ornithosis among "wild" pigeons in Ontario. Canad. Pub. Health Jour. 38:187.

Laubscher, J. H., Wentzien, A. J., and Jordan, C. F.: 1945. Psittacosis in Iowa. Jour. Iowa Med. Soc. 35:44.

Lazarus, A. S., and Meyer, K. F.: 1939. The virus of psittacosis. Jour. Bact. 38:121, 153, and 171.

Leichtenstern, O.: 1899. Über infektiöse Lungenentzündungen und den heutigen Stand der Psittakosis-Frage. Zentralbl. f. allg. Gesundheitspflege. Bonn. 18:241.

Levinthal, W.: 1930. Die Aetiologie der Psittakosis. Klin. Wochenschr. 9:654.

—————: 1935. Recent observations on psittacosis. Lancet 228:1207.

Lillie, R. D.: 1933. The pathology of psittacosis. Nat. Inst. Health, Bul. 161, Washington.

Loizaga, N. S., and Averbach, S.: 1945. Sobre una epidemica de Psittacosis predominio del contagio interhumans. Rev. de med. y. Ciencias Afines 7:298.

Meyer, K. F.: 1934. The heterogeneous infection chains as occupational diseases. Arch. f. Gewerbepath. u. Gewerbehyg. 5:564.

—————: 1935. Psittacosis. Proc. Twelfth Internat. Vet. Cong. 3:182.

—————: 1941a. Phagocytosis and immunity in psittacosis. Schweiz. med. Wochenschr. 71:436.

—————: 1941b. Pigeons and barnyard fowls as possible sources of human psittacosis or ornithosis. Schweiz. med. Wochenschr. 71:1377.

—————: 1942. The ecology of psittacosis and ornithosis. Medicine 21:175.

————— and Eddie, B.: 1933. Latent psittacosis infections in shell parakeets. Proc. Soc. Exper. Biol. and Med. 30:484.

————— and Eddie, B.: 1934a. Psittacosis in the native Australian budgerigars. Proc. Soc. Exper. Biol. and Med. 31:917.

————— and Eddie, B.: 1934b. Latent psittacosis and Salmonella psittacosis infection in South American parrotlets and conures. Science 79:546.

————— and Eddie, B.: 1934c. Eine Psittakoseepidemie in einem Vogelbauer mit Reisvögeln und Sittichen. Berliner tierärztl. Wochenschr. 50:577.

————— and Eddie, B.: 1939a. Psittacosis in importations of psittacine birds from the South American and Australian continents. Jour. Infect. Dis. 65:234.

————— and Eddie, B.: 1939b. The value of the complement fixation test in the diagnosis of psittacosis. Jour. Infect. Dis. 65:225.

————— and Eddie, B.: 1942. Spontaneous ornithosis (psittacosis) in chickens the cause of a human infection. Proc. Soc. Exper. Biol. and Med. 49:522.

————— and Eddie, B.: 1947. The knowledge of human virus infections of animal origin. Jour. Am. Med. Assn. 133:822.

—————, Eddie, B., and Stevens, I. M.: 1935. Recent studies on psittacosis. Am. Jour. Pub. Health 25:571.

—————, Eddie, B., and Yanamura, H.: 1942a. Ornithosis (psittacosis) in pigeons and its relation to human pneumonitis. Proc. Soc. Exper. Biol. and Med. 49:609.

—————, Eddie, B., and Yanamura, H.: 1942b. Active immunization to the *Microbacterium multiforme psittacosis* in parakeets and ricebirds. Jour. Immunol. 44:211.

Morange, A.: 1895. De la psittacose, ou infection spéciale déterminée par des perruches. Thèse de Paris.

Morgan, H. R., and Wiseman, R. W.: 1946. Use of bacteriostatic agents in preparation of seed cultures of psittacosis virus. Proc. Soc. Exper. Biol. and Med. 62:130.

Moshkovsky, S. D.: 1945. The cytotropic agents of infections and the positions of the rickettsiae in the system of chlamydozoa. U spekhi souremennoi biologii (Russian). Rev. and Gen. Theoretical Paper 19:1.

Nigg, C., and Eaton, M. D.: 1944. Isolation from normal mice of a pneumotropic virus which forms elementary bodies. Jour. Exper. Med. 79:497.                                                            .

Nocard, E., and Dobove: 1896. Sur un mémoire de les docteurs Gilbert, A. et Fournier, L. Contribution à l'étude de la psittacose. Bul. Acad. de méd. 36:429.

Olson, B. J., and Larson, C. L.: 1944. An epidemic of a severe pneumonitis in the Bayou Region of Louisiana. IV. Preliminary note on etiology. Pub. Health Rep. 59:1373.

—— and Larson, C. L.: 1945. An epidemic of a severe pneumonitis in the Bayou Region of Louisiana. V. Etiology. Pub. Health Rep. 60:1488.

—— and Treuting, W. L.: 1944. An epidemic of a severe pneumonitis in the Bayou Region of Louisiana. I. Epidemiological study. Pub. Health Rep. 59:1299.

Pacheco, G.: 1931a. Nouvelles recherches sur la psittacose des perroquets. Compt. rend. Soc. de biol. 106:372.

——: 1931b. Nouvelle espèce de Salmonella pathogènes. Différentiation avec l'espèce de Nocard. Comp. rend. Soc. de biol. 106:1018.

—— and Bier, O.: 1930. Epizootie chez les perroquets du Brésil. Compt. rend. Soc. de biol. 105:109.

Parodi, A. S., and Silvetti, L. M.: 1946. La psitacosis en los psitacidos silvestres (*Myiopsitta monacha,* Bodde) de la Republica Argentina. Prensa méd. argent. 33:529.

Pfaffenberg, R.: 1936. Die Psittacosis (Papageienkrankheit) in den Jahren 1931–1935. Epidemiologie, Forschungsergebnisse, Bekämpfung. Ergebn. d. Hyg., Bakt., Immunitätsforsch. u. exper. Therap. 18:251.

Pinero Garcia, P. P.: 1940. Impresiones clínico-epidemiológicas sobre el último paroxismo de psitacosis en la Capital Federal. Prensa méd. argent. 27:2463 and 2514.

Pinkerton, H., and Swank, R. L.: 1940. Recovery of virus morphologically identical with psittacosis from thiamin-deficient pigeons. Proc. Soc. Exper. Biol. and Med. 45:701.

Pollard, M.: 1947. Ornithosis in seashore birds. Proc. Soc. Exper. Biol. and Med. 64:200.

Rake, G., and Jones, H. P.: 1942. Studies on Lymphogranuloma venereum. I. Development of the agent in the yolk sac of the chicken embryo. Jour. Exper. Med. 75:323.

—— and Jones, H. P.: 1944. Studies on Lymphogranuloma venereum. II. The association of specific toxins with agents of the lymphogranuloma-psittacosis group. Jour. Exper. Med. 79:463.

Rasmussen, R. K.: 1938. Ueber eine durch Sturmvögel übertragbare Lungenerkrankung auf den Färöern. Zentralbl. f. Bakt., Abt. I., Orig. 143:89.

Rivers, T. M., and Berry, G. P.: 1932. A laboratory method for the diagnosis of psittacosis in man. Proc. Soc. Exper. Biol. and Med. 29:942.

——, Berry, G. P., and Sprunt, D. H.: 1931. Psittacosis: I. Experimentally induced infections in parrots. Jour. Exper. Med. 54:91.

—— and Schwentker, F. F.: 1934. Vaccination of monkeys and laboratory workers against psittacosis. Jour. Exper. Med. 60:211.

Robinow, C. F., and Bland, J. O. W.: 1938. Application of the Feulgen method to the study of viruses. Nature, London, 142:720.

Rudd, G. V., and Burnet, F. M.: 1941. Intranasal infection of mice with the virus of psittacosis. Austral. Jour. Exper. Biol. and Med. Sci. 19:33.

Ruys, A. C.: 1934. De Verwekker van de Papagaaienziekte. Nedeil. tijdschr. v. geneesk. 78:2095.

St. John, E., and Gordon, F. B.: 1947. Studies on the immunological relationship of the psittacosis lymphogranuloma venereum group of viruses. Jour. Infect. Dis. 80:297.

Smadel, J. E.: 1943. Atypical pneumonia and psittacosis. Jour. Clin. Invest. 22:57.

——, Jackson, E. B., and Harman, J. W.: 1945. A new virus disease of pigeons. I. Recovery of the virus. Jour. Exper. Med. 81:385.

——, Wall, M. J., and Gregg, A.: 1943. An outbreak of psittacosis in pigeons, involving the production of inclusion bodies, and transfer of the disease to man. Jour. Exper. Med. 78:189.

——, Wertman, K., and Reagan, R. L.: 1943. Yolk sac complement fixation antigen for use in psittacosis-lymphogranuloma venereum group of diseases. Proc. Soc. Exper. Biol. and Med. 54:70.

Tomlinson, Jr., T. H.: 1941. An outbreak of psittacosis at the National Zoological Park, Washington, D. C. Pub. Health Rep. 56:1073.

Troup, A. G., Adam, R., and Bedson, S. P.: 1939. An outbreak of psittacosis at the London Zoological Gardens. Brit. Med. Jour. 1:51.

Vervoort, H., and Ruys, A. C.: 1939–1940. The recognition of psittacosis. Antonie van Leeuwenhoek, Nederl. Tijdschr. Hyg. Microbiol. 6:11.

Yanamura, H. Y., and Meyer, K. F.: 1941. Studies on the virus of psittacosis cultivated in vitro. Jour. Infect. Dis. 68:1.

CHAPTER TWENTY-THREE

# AVIAN ENCEPHALOMYELITIS (EPIDEMIC TREMOR)[1]

*By* PETER K. OLITSKY, *The Laboratories of the Rockefeller Institute for Medical Research, New York*

☆    ☆    ☆

**Synonyms.** Epidemic tremor of young chickens, encephalomyelitis in the chicken (Jones, 1932, 1934). The name, infectious avian encephalomyelitis, was given to this malady by Van Roekel, Bullis, and Clarke (1938, 1939). The Committee on Poultry Disease Nomenclature of the American Veterinary Medical Association recommended in 1939 the binomial, avian encephalomyelitis.

**Definition.** Avian encephalomyelitis is an acute viral infection of all standard and cross breeds of chicks and is characterized by a variable incubation, usually prolonged, and by ataxia and tremor especially of the head and neck.

**History.** Jones first observed the disease in May, 1930, in a commercial flock of nine Rhode Island Red, two-week-old chicks. The malady was not encountered again until April, 1931, when birds from another source became involved; and again in the spring of 1932. The three epidemics mentioned occurred in Massachusetts; these were followed in 1932–33 by one or more epidemics which arose in New Hampshire, Massachusetts, Maine, and Connecticut, and which came to Jones's attention. Since then others have reported the disease from widely separated areas.

**Distribution.** Epizootics have broken out not only in the New England States but also in New York, New Jersey, Delaware, Virginia, Indiana, Colorado, Georgia, Tennessee, and Florida; they have also been encountered in Australia (Jungherr and Minard, 1942). In spite of their wide and expanding distribution, poultrymen who have experienced epizootics do not regard them at the present time as a threat to the poultry industry (Van Roekel, personal communication).

**Etiology and properties of the virus.** Jones (1934) stated that the causative agent of the disease should be placed among organisms of the filter-passing group, the writer (1939) confirmed the fact that it is an ultramicroscopic virus, and in collaboration with Bauer (1939) estimated the size of the virus

---

[1] The writer expresses his thanks to the editors of the *Journal of Experimental Medicine* for permission to reproduce illustrations accompanying this article. The cooperation of H. Van Roekel is acknowledged.

particles by means of ultrafiltration through gradocol membranes as 20 to 30 millimicrons in diameter.

The virus has had hitherto but scant study, and a description of its properties, pathogenesis, etc., must necessarily be incomplete. It is of interest, however, that the size of the viral particle is within the same range as that demonstrated for most of the viral encephalitides of man and lower animals. It has been shown that the avian virus has no relation to that of eastern or western equine encephalomyelitis but is one *sui generis* (Olitsky, 1939). Infection of chickens can be brought about by intracerebral injection of brain virus in decimal dilutions from $10^{-1}$ to $10^{-7}$. It is not sedimented at 12,000 r.p.m. for 1 hour; the supernate still yields a high degree of infectivity. The active agent is filtrable through Berkefeld V and N candles and Seitz 1 and 2 disc filters. The active filtrates are free from any visible or cultivable micro-organisms, and no bacteria can be seen in stained film preparations or sections of affected tissues. The infective agent, as contained in nervous tissue, can be preserved in 50 per cent glycerol and by means of lyophilization. Van Roekel (personal communication) found that a brain-saline solution suspension of the virus stored at 38–40° C. for 836 days, when injected into the brain of 1-day-old chicks, induced symptoms and histopathological changes of avian encephalomyelitis.

The virus multiplies in Maitland type cultures of minced whole embryo tissue in vitro in the presence of chick serum (Kligler and Olitsky, 1940). Chick embryos, 1–18 days old, can be infected, but only irregularly so, by various routes of inoculation (Van Roekel et al., 1939; Jungherr and Minard, 1942). Van Roekel (personal communication) found that of 807 eggs inoculated and then incubated, only 149 chicks hatched, of which 71 showed encephalomyelitis, some of which exhibited signs on the day of hatching. Kligler and Olitsky (1940) reported difficulty in infecting chick embryos by allantois and yolk sac routes of inoculation although F. Bang (personal communication) succeeded by introduction of the virus directly into the embryo brain. The virus is present in embryonic remnants of the developed chick, i.e., in unabsorbed egg yolk derived from 2-week-old chicks having the malady in nature (Jungherr and Minard, 1942).

**Pathogenesis.** Jones and Van Roekel and his co-workers have established that intracerebral injection of the virus is followed by uniform and invariable development of the disease. The writer has shown that the regularity of response to cerebral injection depends upon the number of infective doses in the inoculum. After peripheral inoculation, even in young chicks, no such uniformity of results is seen. Small proportions of chicks have been brought down with experimental encephalomyelitis by intraperitoneal, subcutaneous, intradermal, intravenous, intramuscular, and intrasciatic injections. Up to the present time, in only a few trials, feeding virus by gavage

and injecting it intraocularly have proved generally ineffective, although about 10,000 minimal chicken cerebral infective doses have been employed. Van Roekel and his co-workers state that intranasal instillation of brain virus can induce clinical infection, and Jungherr and Minard report that of the peripheral routes used for inoculation, the intravenous is the most successful. Virus has not been detected in the blood during the early stages of clinically apparent experimental disease, nor during the period extending from 1 to 5 days before the first clinical sign was manifest. This weighs against the concept of conveyance of the disease from bird to bird by blood-sucking insects. Finally, the virus—perhaps with slight increment—may persist in the brain for at least 24 days in such birds as have nonprogressive tremor and ataxia (Olitsky, 1939). Investigators are agreed that in the active stages of the malady the brain offers the most constant source of the virus. Van Roekel and his associates (1939) have also recovered it from the spleen and liver.

**Epidemiology.** As stated by Jones and by Van Roekel and his associates, there is a seasonal occurrence of epidemics of the disease during winter and spring months. The age of susceptibility to infection is from the time of hatching to six weeks, with the usual time of onset at one to three weeks of age. Experimentally, the disease can be induced in maturing birds up to at least three months of age (Van Roekel *et al.*, 1939; Kligler and Olitsky, 1940). Jones (1934) holds the opinion that there is no available evidence derived from experiments on mating diseased birds, that transmission of the incitant takes place through the egg; from the work of Van Roekel, it would appear, however, that the virus is egg-borne. Moreover, Jungherr and Minard (1942) conclude that the virus is not only egg-borne but persists in the visceral tissues (especially gonadal) of certain adult birds, and is eliminated *via* the genital or intestinal tracts. The average morbidity rate is about 17 per cent, with limits in different flocks from 0.1 to 50 per cent. The average mortality rate is about 10 per cent, with limits varying from 0 to 65 per cent. Cross breeds are apparently more often involved than standard straight breeds.

During three epidemics, contact experiments in which normal birds were caged with those having the malady failed to induce infection in the former (Jones, 1934). Jungherr and Minard report that the possibility of contact infection is "very limited." Our results are in agreement. Van Roekel, on the contrary, suspects direct contact to be a factor in the spread of the disease. Olitsky and Schlesinger (unpublished data) have attempted in a limited number of experiments to infect young chicks by intracerebral inoculation and simultaneous feeding with filtrates of feces or intestinal contents from (a) normal chickens, (b) experimentally infected 18-day-old chicks during the acute stage, and (c) 50-day-old chickens during the chronic stage of encephalomyelitis. All these attempts failed.

Hatchability of eggs is not influenced by an outbreak of encephalomyelitis. Not every hatch throughout a season becomes infected, and recurrence may take place in flocks in successive or subsequent hatches. Birds that recover from the experimental infection, however, are rendered immune to reinfection and develop serum viral neutralizing antibody to an appreciable degree (Olitsky, 1939). Jungherr and Minard detected neutralizing antibody in the serum also in convalescents of the natural disease; they have failed, however, to show complement-fixation reaction in this disease. We



FIG. 23.1. Infectious avian encephalomyelitis showing ataxia and attempts of bird to right itself by use of wing. (Olitsky, Rockefeller Inst. Med. Res.)

have found, moreover, that chicks which have received active virus but failed to show clinical signs, i.e., those receiving an insufficient number of infective doses, are not rendered resistant thereby.

A limited range of host susceptibility exists for the avian active agent; chicks, turkey poults, ducklings, and young pigeons are susceptible. The ordinary laboratory animals, such as mice, guinea pigs, rabbits, and monkeys, are resistant to the virus introduced intracerebrally.

**Symptomatology.** The incubation period of the experimental disease varies from 5 to about 40 days, with an average period of 9 to 21 days. In nature, outbreaks occur in chicks usually one to two weeks of age. The earliest signs are a dull expression of the eyes and unsteadiness of gait which develops into definite ataxia (Figs. 23.1, 23.2, and 23.3). The legs become weaker, and the chick becomes inactive or sits on its haunches or, when disturbed, walks on its hocks and shanks, frequently falling over on its side. Tremor, especially of the head and neck, is observed, but not in all birds. The weakness of the legs may progress to complete incapacity; general debility increases, and prostration, followed by death, ensues in a few days. Jungherr (1939) states that of histologically positive field cases, 36.9 per cent

showed ataxia, 18.3 per cent, tremor, and 35 per cent, both, and 9.2 per cent, no clinical signs. It is Van Roekel's opinion that death may result either because the chicks, while in a helpless state, are trampled on by the more lively members of a flock, or from inanition due to inability to get to sources of food. It has been our experience, as well, that life can be extended or even spared with proper care of disabled birds.

In other cases, tremulous birds without much ataxia may continue in this state with a general degree of health compatible with normal functioning,



Fig. 23.2. Infectious avian encephalomyelitis; incubation 13 days, ill 2 days. (Olitsky, Rockefeller Inst. Med. Res.)

and in still others the nervous signs may wholly disappear in time. It should be emphasized that the disease may be overlooked in cases with almost imperceptible clinical signs. In some birds the only sign may be slight, almost unrecognizable swaying in an otherwise healthy condition, and this sign can develop even after a three-week incubation period; diagnosis is then confirmed by the characteristic lesions in the central nervous system and viscera (v. infra).

**Pathology.** The lesions are only microscopic. One observes no definite meningeal reaction except that the vessels therein show perivascular infiltration. The lesion most commonly met with, and the most striking, is neuronal degeneration, involving the cells throughout the central nervous system, but more extensively in the pons-medulla and in the anterior horn cells of the spinal cord, especially in the lumbosacral enlargement. The neuron first

becomes rounded in outline and enlarged or swollen; the nucleus also increases in size. The next stage consists of an eccentric placement of the nucleus, which may reach the very limits of the cell membrane. At the same time, there is clearing of the cytoplasm of Nissl substance. There is at first only a halo of Nissl bodies about the periphery of the perikaryon; later, even this thin line of granulation disappears, along with the nucleus, so that finally the perikaryon is stained homogeneously pink to red (in sections colored by eosin- or phloxin-methylene blue or by Giemsa's method). Certain of the



FIG. 23.3. Infectious avian encephalomyelitis showing characteristic ataxia and squatting. (Olitsky, Rockefeller Inst. Med. Res.)

cells in this latter condition exhibit a bright red, round mass, which is not the nucleus or nucleolus, about 2 to 5μ in diameter, and which stands out in sharp contrast to the reddish, even or smooth background. In advanced stages, only faint pink shadows of the attacked neurons are left, and now and again the cell completely disappears. It is noteworthy that in early cases of the affection there may occasionally be no other sign of involvement of the nervous tissue, especially no inflammation or secondary reactions. This is more likely to happen in the anterior horns of the cord. The process resembles Nissl's degeneration, or axon reaction. In the early stages of the malady, most of the Purkinje cells are, as a rule, found to be well preserved. Here and there some cells show degeneration, and in exceptional cases, especially late in the affection, considerable destruction of them occurs.

Another change in the nervous tissue is the perivascular reaction which may reach an extraordinary degree. This reaction pervades the entire brain, especially the cortex, pons-medulla, and cerebellum (Figs. 23.4 and 23.5). The perivascular infiltration is mostly by lymphocytes, with an occasional large monocytic cell, and the dense collar may comprise a depth of ten or more rows of such cells. Jungherr (1939) points out that there occurs mural lymphocytic infiltration of capillaries which also exhibit considerable endothelial hyperplasia. In chronic (quiescent) ataxic and tremulous birds, some



FIG. 23.4. Perivascular lesion in cerebellum and loss of Purkinje cells. Incubation period 14 days; ataxic for 22 days. ×125. (Olitsky, Jour. Exper. Med.)

indications of neuronophagia by glial elements are visible, also small accumulations of such cells, and rarely satellitosis. Jungherr reports that the histopathological changes characteristic of the encephalomyelitis could be found in 9.2 per cent of a series of birds that were normal looking (v. supra) but were in flocks among which epizootics arose. In our experience with the experimental disease, lesions were observed only in animals which exhibited signs of the malady, even those of the mildest type.

As is known, the chicken utilizes for its lymphatic glandular system small islands of lymphoid tissue, sometimes located in the parenchyma and often associated with blood vessels. They are found in all organs, including the gizzard, and in the brain as well, where they are localized in the choroid plexus. Under the influence of avian virus infection these areas become markedly hyperplastic and show, apart from lymphocytes, few monocytes and myelocytes and some cellular debris. Practically all organs are so involved, but especially the liver (Fig. 23.6), the pancreas, and the spleen. In the

heart, the lymphocytic cells may arrange themselves longitudinally between muscle fibers, and probably this arrangement is the consequence of the vascular distribution of the lymphoid tissue.

No definite inclusion bodies can be detected, and film preparations of affected tissue or centrifugalized sediments fail to reveal elementary bodies.

**Diagnosis.** Attention has already been drawn to the fact that on the one hand the disease may pass clinical recognition, and on the other, it may be confused with other maladies of nervous type. From *equine encephalomye-*
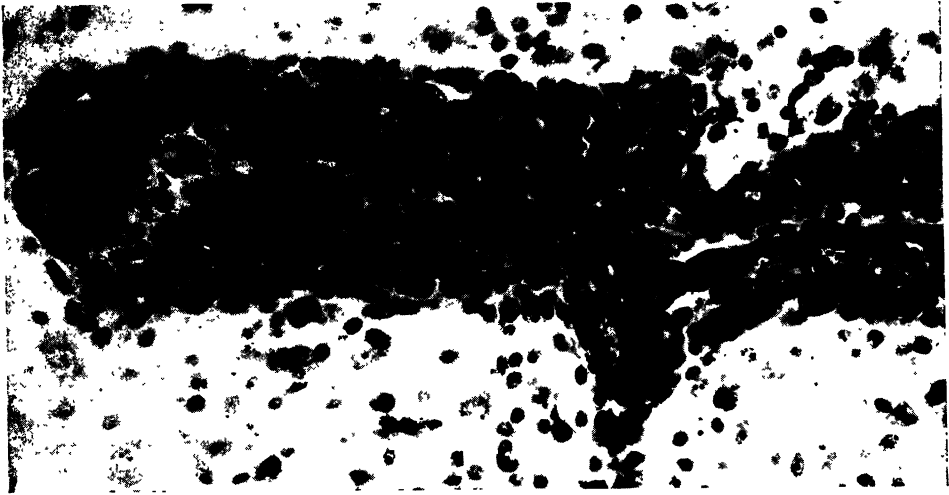


Fig. 23.5. Details of perivascular lesion in cerebellum. ×500. (Olitsky, Jour. Exper. Med.)

*litis* infection, differentiation can be made chiefly through inoculation of mice and other laboratory animals and by serum-neutralization tests. From *avian pneumoencephalitis* (or the nervous form of *Newcastle virus disease*) the differentiation is by specific serum-neutralization tests and by the absence here of the characteristic changes in the nervous tissue and the lymphoid tissue hyperplasia noted in the encephalomyelitis, which, in turn, does not exhibit lesions in the respiratory tract. Affections due to *nutritional disturbances,* such as rickets, nutritional encephalomalacia, and the condition described by Dunlap, i.e., ataxia accompanied by changes in the kidney and proventriculus, should be borne in mind in making differential diagnoses. The transmissibility of avian encephalomyelitis, its distinctive pathology, and absence of nutritional factors in its causation should aid in a decision. *Salmonellosis* of chickens and *coccidiosis* can be determined by bacteriological study. *Cage paralysis* of birds which arises when they are kept in close quarters for long periods of time offers a problem for differentiation, but this condition is not transmissible. Finally, differentiation must be made from

*lymphomatosis gallinarum.* Encephalomyelitis occurs in the absence of tumor formation, enlargement of the dorsal root ganglia, peripheral neuritis, iritis, and involvement of the intestinal tract.

In sum, the diagnostic histopathological signs are the gliosis, the lymphocytic perivascular infiltration and the axon-type of neuronal degeneration in the central nervous system, and the hyperplasia of the visceral lymphoid follicles; Jungherr (1939) adds the special sign of lymphoid hyperplasia in the stomach muscles. The diagnostic serological test consists of determination
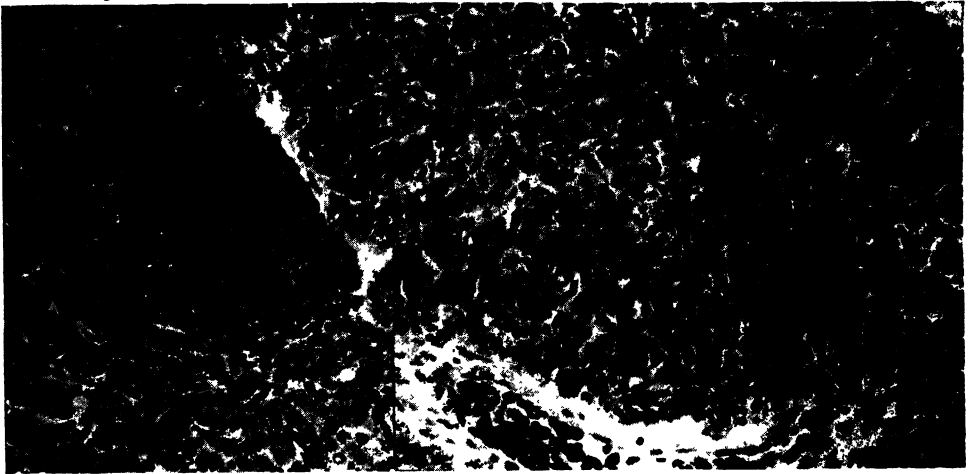


FIG. 23.6. Hyperplasia of the lymphoid islands of liver. ×275. (Olitsky, Jour. Exper. Med.)

of the presence of specific viral neutralization antibody in the blood serum.

**Prevention and treatment.** Van Roekel and Jungherr and co-workers offer little encouragement at the present time for the control of the disease. They believe that infected birds should be culled and disposed of. They advise good husbandry practice and hygiene and condemn any restocking of older, infected flocks with day-old chicks. When breeders are saddled with epidemics arising in repeated waves, relief from this burden, then, is to clear out the stock and start anew.

Olitsky and Schlesinger have shown that two or four doses of formalin-inactivated virus given subcutaneously at 7-day intervals give rise to serum-neutralizing antibody. Undiluted serum neutralized 10–100 cerebral m.i.d. when the test was made about one month after the beginning of vaccination. The vaccinated birds failed, however, to show any resistance to an intra-cerebral test dose of virus given at that time. Whether this degree of antibody formation would protect against a peripheral inoculation—were such a peripheral route available for this method of testing resistance—is left for future study. (The difference in degree of antibody production required for

prevention of infection by different routes with a neurotropic virus is described by Morgan, Schlesinger, and Olitsky, 1942.)

## REFERENCES

Jones, E. E.: 1932. An encephalomyelitis in the chicken. Science 76:331.

.———: 1934. Epidemic tremor, an encephalomyelitis affecting young chickens. Jour. Exper. Med. 59:781.

Jungherr, E.: 1939. Pathology of spontaneous and experimental cases of epidemic tremor. Poultry Sci. 18:406.

——— and Minard, E. L.: 1942. The present status of avian encephalomyelitis. Jour. Am. Vet. Med. Assn. 100:38.

Kligler, I. J., and Olitsky, P. K.: 1940. Experiments on the cultivation of virus of infectious avian encephalomyelitis. Proc. Soc. Exper. Biol. and Med. 43:680.

Morgan, I. M., Schlesinger, R. W., and Olitsky, P. K.: 1942. Induced resistance of the central nervous system to experimental infection with equine encephalomyelitis virus. I. Neutralizing antibody in the central nervous system in relation to cerebral resistance. Jour. Exper. Med. 76:357.

Olitsky, P. K.: 1939. Experimental studies on the virus of infectious avian encephalomyelitis. Jour. Exper. Med. 70:565.

——— and Bauer, J. H.: 1939. Ultrafiltration of the virus of infectious avian encephalomyelitis, Proc. Soc. Exper. Biol. and Med. 42:634.

Van Roekel, H., Bullis, K. L., and Clarke, M. K.: 1938. Preliminary report on infectious avian encephalomyelitis. Jour. Am. Vet. Med. Assn. 93:372.

———, Bullis, K. L., and Clarke, M. K.: 1939. Infectious avian encephalomyclitis. Vet. Med. 34:754–55.

# EQUINE ENCEPHALOMYELITIS VIRUS IN BIRDS

By L. T. Giltner, *Pathological Division, Bureau of Animal Industry,*
*United States Department of Agriculture, Washington, D. C.*

☆   ☆   ☆

During the last decade considerable investigational work has been done on infectious equine encephalomyelitis with particular reference to the search for hosts other than the horse. Giltner and Shahan (1933) found that pigeons could be readily infected by intracerebral inoculation with the western type virus. The inoculated birds developed symptoms of general weakness, ataxia, and marked tremors on the third day and died on the third or fourth day in a state of complete paralysis. Those authors (Giltner and Shahan, 1936), as well as Ten Broeck (1938) and Howitt (1939a), have shown that chickens are susceptible to artificial infection. Remlinger and Bailly (1936a, b) successfully infected by subdural inoculation with western type virus (Argentine) the graylag goose (*Anser cinereus*), the hawk (*Circus rufus*), European blackbird (*Turdus merula*), the tawny vulture (*Vultur fulvus* Briss), the white stork (*Ciconia alba*), and the common mallard duck (*Anas boschas*). Ten Broeck (1938) and Howitt (1939a) have reported experimental infection of turkeys, and Shahan, Giltner, and Schoening (1938) have found the guinea fowl (*Numida meleagris*) to be susceptible to both eastern and western type viruses by intracerebral inoculation. Tyzzer, Sellards, and Bennett (1938) produced typical encephalomyelitis in adult quail by intracerebral inoculation with eastern type virus. English sparrows (*Passer domesticus*) were found very susceptible to intracerebral infection with eastern type virus by Van Roekel and Clarke (1939). Davis (1940) reports that sparrows are susceptible to either intracerebral or subcutaneous inoculation with eastern virus, and that the mosquito, *Aedes sollicitans,* is capable of infecting these birds with eastern virus. He also reports that the cowbird succumbs to either subcutaneous or intracerebral inoculation. The mosquito, *Aedes vexans,* also transmits a fatal infection to these birds. Howitt (1940) found that the valley quail (*Lophortyx californica*) is definitely susceptible to the virus of equine encephalomyelitis. Rosenbusch (1939) found a hawk (*Milvajo chimango*) to be susceptible to the virus. Rosenbusch (1939) inoculated three large sea gulls (*Larus belcheri*) and eight smaller sea gulls (*Larus dominicanus*), but the birds did not prove to be susceptible.

The first outbreak of the natural disease in birds was recorded by Tyzzer, Sellards, and Bennett (1938) who encountered fatal infection in ring-necked pheasants in Connecticut. In the same year Fothergill and Dingle (1938) found the natural infection in a pigeon in Massachusetts. Van Roekel and Clarke (1939) and Beaudette (1939) studied natural outbreaks in pheasants in New Jersey. The latter investigator recovered eastern type virus from three distinct outbreaks and reported that the time of the appearance of the



Fig. 24.1. Heart of chicken showing great distention of right and left ventricles. (Tyzzer and Sellards, Am. Jour. Hyg.)

disease in birds paralleled that in horses in the general vicinity. The disease appears to have begun in late August and continued up to early November. The symptoms reported include paralysis of the legs, head drawn over back, staggering, and failure to eat or drink. Death occurred in 1 or 2 days or the bird sometimes recovered slowly.

Although the mode of natural transmission has not been definitely determined, there is experimental evidence which strongly points to mosquitoes as vectors of the disease. Davis (1940) found that the mosquitoes (*Aedes atropalpus, A. vexans, A. sollicitans, A. cantator, A. triseriatus,* and *A. aegypti*) were capable of transmitting the eastern equine encephalomyelitis virus from infected birds to normal animals after a 9-day incubation period. Mosquitoes fed on infected birds transmitted virus to mice and birds; those fed on mice transmitted it to birds, mice, and guinea pigs.

Ten Broeck' (1939) showed that the American egret is susceptible to the infection. The Gambel sparrow, the junco, and the thrasher were found susceptible by Howitt (1939b). At the same Congress, Syverton (1939) reported that the western burrowing owl is quite susceptible.

Cox, Jellison, and Hughes (1941) recovered the virus of western equine encephalomyelitis from the brain and spleen of a prairie chicken (*Tympanuchus cupido americanus* Reichenbach). The bird was shot August 27,
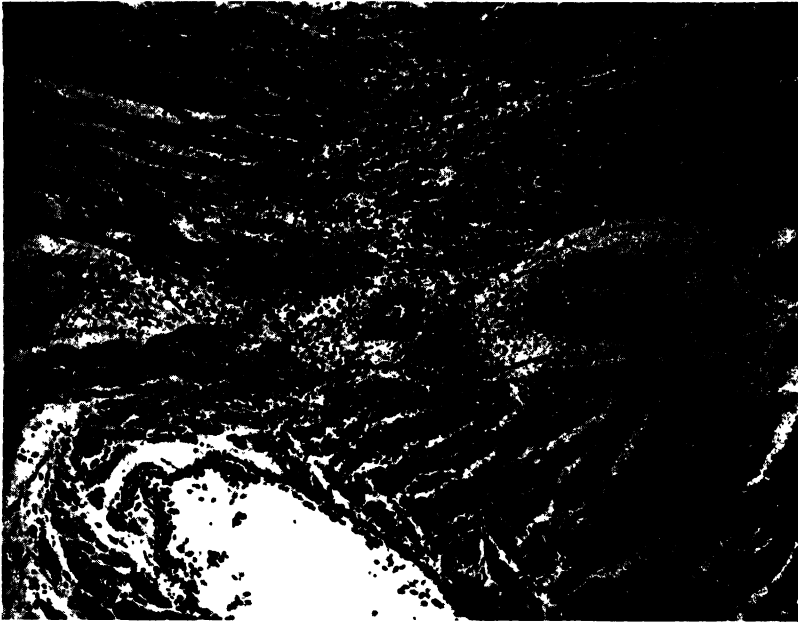


FIG. 24.2. Myocardium of chick showing edema around vessels and diffuse infiltration. ×80. (Tyzzer and Sellards, Am. Jour. Hyg.)

1941, near Rugby, North Dakota, while human cases were occurring in the vicinity.

Sellards and associates (1941) produced a fatal paralysis in adult pheasants by intramuscular injection of virus recovered from pheasants which had died in Connecticut. The virus showed the characteristics of eastern equine encephalomyelitis virus. They also killed very young chicks within 48 hours by subcutaneous injection. Intracerebral inoculation of young hens failed to produce symptoms, but their sera developed protective properties.

Tyzzer and Sellards (1941) inoculated very young Rhode Island Red chickens with a pheasant strain of eastern equine encephalomyelitis virus. Some of the birds died in from 2 to 4 days without showing notable symptoms, while others showed inactivity, weakness, ruffled plumage, or stupor without paralysis. Some manifested no symptoms. Pathological changes were found

in the brain, heart, gizzard musculature, and sometimes in the liver. The heart was distended, the right ventricle showing the greatest dilatation (Fig. 24.1). The histologic changes consisted of a well-developed myocarditis (Fig. 24.2).

The central nervous system frequently revealed slight perivascular infiltrations (Fig. 24.3); occasionally extensive degenerative changes were noted. Focal infiltration was frequently found in the smooth muscle of the gizzard. Degenerative changes were also seen in the liver in several instances. In
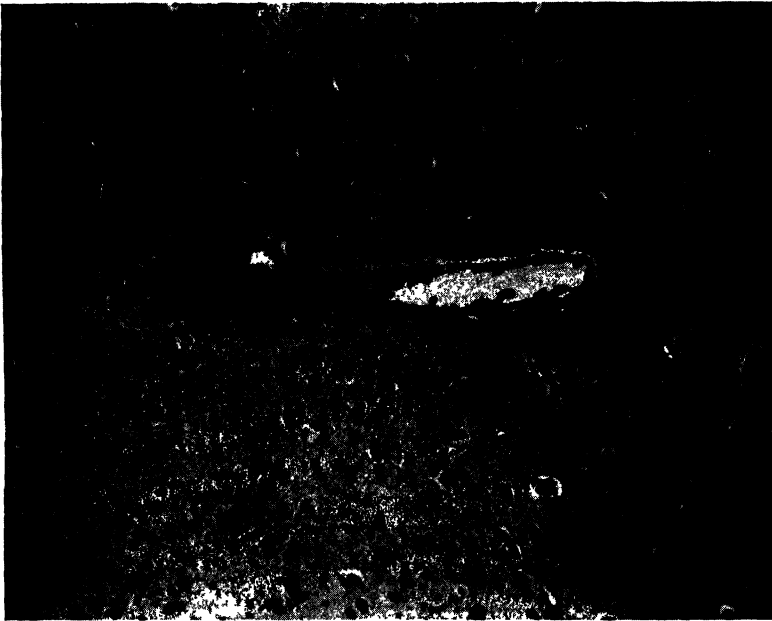


Fig. 24.3. Cerebral blood vessel with localized inflammation in chick. ×160. (Tyzzer and Sellards, Am. Jour. Hyg.)

young chickens the cardiac lesions were the most striking, overshadowing the histologic changes seen in the central nervous system. The distribution of the histopathology of equine encephalomyelitis as seen in the pheasant, quail, and young chicken is in distinct contrast to that of mammals.

Mitchell and Walker (1941) studied the effect of western equine encephalomyelitis virus in two geese. "Two days after inoculation both showed evidence of illness characterized by inability to stand, loss of appetite, rapid dehydration from which they gradually recovered except for a stunted growth. Suddenly, 46 days after inoculation, goose No. 1 showed symptoms suggestive of central nervous system involvement. The bird was at first weak, then unable to stand, the tone of voice altered, later became hoarse and finally inaudible. Progressive weakness was evident . . ." The goose died 2 days

following the appearance of the latter symptoms. Goose No. 2 presented similar syndromes and died 56 days after inoculation.

Blood and brain of the geese were capable of infecting guinea pigs, but the virus was unable to permanently adapt itself to guinea pigs. Many of the surviving guinea pigs were found immune when subsequently challenged with the western type of virus.

Sulkin (1945) reported the recovery of western type virus from chicken mites (*Dermanyssus gallinae*) found on a ranch in an area in Texas where an outbreak of equine encephalomyelitis had occurred in horses as well as in man during the summer and fall of 1944. The serum from chickens on the same ranch neutralized western type equine virus.

Hammon and Reeves (1946) showed that chickens inoculated subcutaneously with western equine encephalomyelitis virus had virus in the blood between the twelfth and forty-eighth hour but showed no signs of illness. Minimal infective doses of virus for chickens led to multiplication of virus so that it was detectable in the serum in $10^{-4}$ dilution. Virus was not found to persist in any organ of the chicken for more than 3 days after inoculation and usually did not persist over 2 days. Antibodies were present in the blood within at least 15 days after inoculation.

Chickens may serve as sources of infection for mosquitoes or other blood-sucking ectoparasites for short periods after the infecting bite of a similar invertebrate vector, but there is no evidence that chickens serve as latent carriers of the virus.

## REFERENCES

Beaudette, F. R.: 1939. Equine encephalomyelitis in avian hosts. Proc. 43rd Annual Meeting of the U. S. Livestock Sanitary Assn., p. 185.

Cox, H. R., Jellison, W. J., and Hughes, L. E.: 1941. Isolation of western equine encephalomyelitis virus from a naturally infected prairie chicken. U. S. Public Health Rep. 56:1905.

Davis, W. A.: 1940. A study of birds and mosquitoes as hosts for the virus of eastern equine encephalomyelitis. Am. Jour. Hyg. 32 (Sec. C) :45.

Fothergill, L. D., and Dingle, J. H.: 1938. A fatal disease of pigeons caused by the virus of the eastern variety of equine encephalomyelitis. Science 88:549.

Giltner, L. T., and Shahan, M. S.: 1933. Transmission of infectious equine encephalomyelitis in mammals and birds. Science 78:63.

———— and Shahan, M. S.: 1936. The present status of infectious equine encephalomyelitis in the United States. Jour. Am. Vet. Med. Assn. 88:363.

Hammon, W. M., and Reeves, W. C.: 1946. Western equine encephalomyelitis virus in the blood of experimentally inoculated chickens. Jour. Exper. Med. 83:163.

Howitt, B. F.: 1939a. The virus of equine encephalomyelitis. Proc. Third Internat. Cong. for Microbiol., New York, p. 92.

————: 1939b. Equine encephalomyelitis; its relationship to man and animals in California. Proc. Third Internat. Cong. for Microbiol., p. 302.

————: 1940. Comparative susceptibility of wild and domestic birds and animals to the western virus of equine encephalomyelitis (Br. strain) in California. Jour. Infect. Dis. 67:177.

Mitchell, C. A., and Walker, R. V. L.: 1941. Studies in equine encephalomyelitis. Susceptibility of some mammals and birds. Canad. Jour. Comp. Med. 5:314.

Remlinger, P., and Bailly, J.: 1936a. Transmission de l'encephalomyélite Argentine des équides à l'oie, au canard, à la buse et au merle. Compt. rend. Soc. de biol. 121:146.

————— and Bailly, J.: 1936b. Transmission de l'encéphalo-myélite des équides (virus Californien) au vautour fauve (*Vultur fulvus* Briss). Compt. rend. Soc. de biol. 123:562.

Rosenbusch, F.: 1939. Equine encephalomyelitis in the Argentine in its experimental aspects. Proc. 6th Pacific Sci. Cong. Vol. 5:209. (Univ. of Calif. Press, Berkeley and Los Angeles, 1942.)

Sellards, A. W., Tyzzer, E. E., and Bennett, B. L.: 1941. The infection of birds with the virus of equine encephalomyelitis. Am. Jour. Hyg. 33 (Sec. B) :63.

Shahan, M. S., Giltner, L. T., and Schoening, H. W.: 1938. A review of the 1938 outbreak of infectious equine encephalomyelitis in the United States. Proc. 42nd Annual Meeting of U. S. Livestock Sanitary Assn., p. 145.

Sulkin, S. E.: 1945. Recovery of equine encephalomyelitis virus (western type) from chicken mites. Science 101:381.

Syverton, J. T.: 1939. Discussion. Proc. Third Internat. Cong. for Microbiol., p. 306.

Ten Broeck, C.: 1938. Birds as possible carriers of the virus of equine encephalomyelitis. Arch. Path. 25:759.

———: 1939. Transmission of equine encephalomyelitis. Proc. Third Internat. Cong. for Microbiol., New York, p. 300.

Tyzzer, E. E., and Sellards, A. W.: 1941. The pathology of equine encephalomyelitis in young chickens. Am. Jour. Hyg. 33 (Sec. B) :69.

———, Sellards, A. W., and Bennett, B. L.: 1938. The occurrence in nature of "equine encephalomyelitis" in the ring-necked pheasant. Science 88:505.

Van Roekel, H., and Clarke, M. K.: 1939. Equine encephalomyelitis virus (eastern type) isolated from ring-necked pheasants. Jour. Am. Vet. Med. Assn. 94:466.

# FOWL POX

*By* CHAS. H. CUNNINGHAM, *Department of Bacteriology and Public Health,
Michigan State College, East Lansing, Michigan*

☆    ☆    ☆

**Synonyms.** Chicken pox, avian pox, bird pox, contagious epithelioma, sorehead, avian molluscum, avian diphtheria, bird pox diphtheria, canker, geflügelpocken (German), variole aviaire (French), viruela aviar (Spanish), difteria aviar (Spanish), bouba (Portugese).

**History.** Fowl pox has been observed in avian species from time immemorial. The identity of the disease and its possible relationship to small-pox and diphtheria was the subject of considerable concern during the ravages of these diseases in the human population. The presence of fowl pox assumed an alarming significance because of the natural conjecture that birds affected with fowl pox constituted a reservoir of variola and diphtheria for man. An excellent review of the disease is presented by Goodpasture (1928). Concern over the possible biological relationship of fowl pox to variola and vaccinia stimulated research work as to the etiology of the diseases and their classification.

**Etiology.** The causal agent of fowl pox is a filtrable virus as was first demonstrated by Marx and Sticker (1902). Later, Carnwarth (1908) and others showed that the virus is responsible for both the cutaneous and diphtheritic forms of the disease. There appear to be at least four different viruses or strains of virus causing pox among birds: fowl pox virus, pigeon pox virus, canary pox virus, and turkey pox virus. Each virus is infective for its homologous host and in some instances for heterologous hosts. Fowl pox virus causes pox principally among chickens and turkeys. Pigeon pox virus causes pox among pigeons and canary pox virus causes pox among canaries. Turkey pox virus causes pox among turkeys.

The identification of the common etiology of the pox viruses has been attempted in the past by passage of the viruses through homologous and heterologous hosts and using the criteria of immunogenesis and pathogenesis for the interpretation of the results. In many investigations on the pox viruses, quantitative determinations of the virus content of the vaccines were not taken into consideration, and it seems obvious that an accurate assessment of the antigenic properties of such preparations would be impossible.

The present knowledge of avian pox does not allow an adequate classification of the pox viruses without reference to host origin. Some avian pox viruses may be mono-, bi-, or tri-pathogenic with respect to transmission to certain species of birds.

Gallagher (1917) described an outbreak of fowl pox in quail transmissible to chickens. Ward and Gallagher (1920) stated that pox occurs naturally among geese, ducks, and guinea fowl, and that pheasants and various wild birds are also susceptible. Te Hennepe (1926) reported that of 268 ducks examined by him during 1924, 17 were affected with mouth lesions and 14 with cutaneous lesions of fowl pox. Later, te Hennepe (1927) did not observe fowl pox infection in 304 ducks examined during 1925–26. Doyle and Minett (1927) were unable to infect ducks or a seagull with fowl pox virus. Pigeons are generally resistant to infection with fowl pox virus, but Doyle and Minett were able to adapt a strain of the virus to the pigeon by frequent serial passage.

Irons (1934) studied the immunological relationship of several strains of fowl, turkey, and pigeon pox viruses and suggested that some strains of avian pox may be rendered "bi-pathogenic" by passage through a series of heterologous hosts, e.g., fowl pox virus passaged through pigeons. All of the pigeon pox strains studied were found to be infective for chickens. Negative results were obtained in attempting to infect pigeons with a turkey virus. Crows, hawks, owls, ducks, guinea fowls, starlings, and several other species were refractory to the fowl and pigeon strains of virus. Chickens were susceptible but pigeons were refractory to turkey virus. One strain of pigeon pox virus proved infectious for the English sparrow and certain related species. After a single passage in chickens the virus of pigeon pox was greatly attenuated for the pigeon. Repeated passage of the pigeon pox virus in chickens, with one possible exception, failed to destroy the infectivity of the virus for pigeons. One strain of fowl pox virus was transmissible with gradually increasing virulence in pigeons, but was temporarily attenuated for chickens. Two other strains of fowl pox virus were noninfectious for pigeons.

Beach (1939) classified the avian pox diseases according to the host— fowl, turkey, pigeon, and canary—with a list of the species in which it occurs or to which it has been transmitted.

Syverton and Cowan (1944) reported for the first time the recovery of fowl pox virus from a natural outbreak of the disease in the sooty grouse. They state that although avian pox is reported to have occurred under natural or experimental conditions in a wide variety of birds, more conclusive evidence appears to be limited to the chicken, pigeon, turkey, guinea fowl, canary, partridge, quail, and pheasant.

The virus of turkey pox usually has been considered to be the same as that of fowl pox, but the results obtained by some investigators indicate that there

are certain strain differences between viruses causing pox in these two species. Brunett (1934) studied the immunological and pathological relationship between a strain of pox virus obtained from a natural case of the disease in turkeys and a strain of fowl pox virus. No differences were observed in the effect of the two viruses on turkeys, chickens, and pigeons except that the disease was of a longer duration in turkeys and chickens inoculated with fowl pox virus, as compared to inoculation with turkey pox virus. Immunity studies showed that turkeys and chickens inoculated with turkey pox virus or fowl pox virus were immune to subsequent inoculations with these viruses. Pigeon pox virus produced a severe reaction on turkeys, but it failed to produce any immunity to turkey or fowl pox viruses.

Brandly and Dunlap (1938) inoculated chickens with a strain of pox virus obtained from a natural case of the disease in turkeys and observed only a mild cutaneous reaction. No infection was produced at the fifth passage of the virus in chickens.

Beaudette and Hudson (1941) reported that a strain of turkey pox virus studied by them produced a more severe local and systemic reaction and a higher percentage of secondary head lesions in chickens than the fowl pox virus. The lesions consisted of a typical scab of considerable thickness as compared to the atypical scab formation and decreased virulence of the turkey strain studied by Brandly and Dunlap (1938). The turkey virus was less lethal for embryonated chicken eggs than was the fowl pox strain. In cross-immunity tests, Brandly and Dunlap used canary, pigeon, turkey, and fowl pox viruses for a study of the immunological relationships of these viruses and concluded " . . . . that canary virus immunizes against itself and against pigeon virus to a high degree. The canary virus seems to produce no immunity against turkey virus but apparently a slight immunity to the fowl virus. Similarly, pigeon virus protects against itself and canary virus but does not give complete protection against turkey and fowl viruses. Turkey and fowl viruses give almost complete protection against the four viruses used."

Kikuth and Gollub (1932) found in their work on bird malaria a virus capable of producing a highly fatal disease of canaries. This virus has been the subject of controversy concerning its relationship to the avian pox viruses.

Burnet (1933a) concluded that Kikuth's virus was a member of the bird pox group. The virus was not pathogenic for day-old chicks, half-grown fowls, pigeons, and parrots (budgerigars). Sparrows were susceptible to infection.

Burnet and Lush (1936) showed by utilization of the embryonated chicken egg technic for quantitative determination of viral activity and antibody content of serum that fowl pox and Kikuth's strain of canary pox, although not identical, are serologically related. They state: "The demonstration of the close serological relationship of these two strains of virus, which

produce superficially very distinct lesions on two different host species, could hardly have been made by any other technic, and certainly not with the same degree of accuracy. If the canary-pox and fowl-pox are serologically almost identical, it is more than likely that all bird-pox strains are similarly related. The advantages of having a common susceptible organism for the whole group are so obvious that it is hoped that investigators having access to material . . . . will explore the possibilities of the method used in the present study."

Burnet (1933b) was able to infect canaries with pox material collected from a spontaneous outbreak in wild sparrows. The disease in the canaries was similar to that produced by Kikuth's canary virus, but the difficulties in filtration of the virus and the agglutinative tendencies of the virus particles resembled fowl pox virus.

McGaughey and Burnet (1945) studied three cases of avian pox in wild sparrows and concluded that the virus resembled canary pox in causing a fatal disease in sparrows and canaries and localized lesions in fowls and pigeons.

The strain of canary pox virus studied by Reis and Nóbrega (1937) was "tri-pathogenic" as it infected chickens, pigeons, and canaries. The virus did not dissociate into mono-pathogenic strains after serial passage through chicks, pigeons, and canaries. Typical inclusion bodies were found in the infected birds, and those which recovered developed immunity against the homologous virus as well as fowl and pigeon pox viruses. Birds vaccinated with either fowl pox or pigeon pox viruses developed immunity against the canary pox strain. The canaries from which the strain was isolated suffered from a severe pox disease in which the mortality was 98 per cent.

Grosso and Prieto (1939) have reported a "tri-pathogenic" canary virus which produced only a mild, transitory, localized lesion in chicks and pigeons.

Antoniotti and Romat (1940) studied a canary pox virus which was pathogenic only for canaries.

Durant and McDougle (1938) conducted studies of the immunological relationship of canary pox virus to fowl pox virus with the following conclusions: "Chickens, turkeys, and quail when inoculated with canary pox virus, though fairly typical gross lesions are produced, later when inoculated with fowl-pox virus will develop typical lesions, indicating that no immunity to the fowl-pox virus had been developed by exposure to the canary-pox. This would seem to indicate that canary-pox virus is a different type of virus with a different degree of virulence for other bird species than the canary."

Coulston and Manwell (1941) reported that canary pox virus was found to be infective for English and song sparrows but not for cowbirds, starlings, or chickens. Canaries which had been vaccinated with the virus attenuated through long storage did develop an immunity which protected them to

some degree, but not completely, against exposure to fully virulent virus. Vaccination with the virulent virus was always fatal.

**Pathology.** Detailed descriptions of the pathological processes in fowl pox are reported by Goodpasture (1928) and Hutyra, Marek, and Manninger (1938).

Fowl pox is characterized by the appearance of cutaneous eruptions or wartlike nodules on the unfeathered parts of fowl and diphtheritic membranes of the mouth. The lesions are observed particularly about the head region, but they may also appear on the legs and feet, around the cloacal aperture, and under the wings. The characteristic lesion of the cutaneous form of fowl pox is a local epithelial hyperplasia involving both epidermis and underlying feather follicles with the formation of nodules. The cutaneous nodules may be very numerous or few in number, and they do not necessarily erupt at the same time. At first, the nodules appear as small, whitish foci which rapidly increase in size and become yellowish in color as they develop. In some instances, closely adjoining lesions may coalesce, and the larger developing lesions are rough, and gray or dark brown in color. After about two weeks of development, sometimes sooner, the lesions may show areas of inflammation at their base and become hemorrhagic. The lesion then undergoes a process of desiccation and scab formation which may last for another week or possibly two weeks. In uncomplicated cases the process ends with desquamation of the degenerated parts of the epithelial layer. If the desiccated scab is removed in the meantime, a moist, sero-purulent exudate is found underneath covering a bleeding, granulating surface. When the scab drops off, a smooth scar may be present, although in milder lesions there may be no noticeable evidence of scar tissue. The specific process is often modified by the invasion of bacteria which propagate in the degenerated epithelium and may reach the deeper layers of the mucous membrane where they cause suppurative or necrotic processes with the formation of fibrinous deposits.

The eruptions on the mucous membranes are white, opaque, slightly elevated nodules. These processes rapidly increase in size, often coalescing to become a yellowish, cheesy, necrotic material with the appearance of a pseudomembrane. When these pseudomembranes are removed they leave bleeding erosions. The invasion by contaminating bacteria aggravates the diphtheritic form of the disease. The inflammatory process may extend from the mouth region into the sinuses, particularly the infraorbital sinuses, resulting in a tumor-like swelling, and may extend into the pharynx, resulting in respiratory disturbances.

Stafseth (1931) investigated a natural outbreak of pox in pigeons and reported that infected pigeons showed typical pox lesions on various parts of

the body. Most of the scabs were found on the feet, legs, and near the base of the beak. A few pigeons showed cankers in the mouth. Hutyra, Marek, and Manninger (1938) reported that in pigeons the pox lesions develop chiefly on the borders of the eyelids, the angles of the mouth, and on the feet.

The characteristics of the canary disease of Kikuth and Gollub (1932) show little resemblance to fowl pox, but Burnet (1933a) and Burnet and Lush (1936) presented evidence that the virus of the disease was a member of the bird pox group and classified it as canary pox. Intramuscular injection of the virus produced an inflammatory necrotic local lesion and a generalized infection throughout the blood. The post-mortem appearance resembled that of a subacute bacterial cellulitis. Hemorrhages under the serous membranes, edema of the lungs, and pericarditis were observed. The difference between the pathological manifestations of fowl pox infection and Kikuth's canary pox infection, according to Burnet (1933a), was the capacity of the canary pox virus to multiply within the cytoplasm of the cells derived from all three primary germinal layers. Mononuclear cells were infected and spread the infection in many respects comparable to the spread of a pyogenic bacterial infection.

Durant and McDougle (1938) observed that in canaries the external pox lesions affected the entire body. The lesions were discrete, usually round, white tinged with yellow containing a serous puslike material. Scab formation was not as distinct as in chickens, although there was a tendency toward this condition in the advanced stages of the disease. Cheesy exudates were found in the commissures of the mouth and the entrance to the larynx.

Canary pox, as reported by Coulston and Manwell (1941), may be manifested by lesions about the margin of the epithelium of the eye and elsewhere on the head, and by lesions of the toes and legs.

In common with many virus diseases, the significant feature of the histological picture of fowl pox is the presence of intracytoplasmic inclusion bodies in the affected epithelial cells (Fig. 25.1). Goodpasture (1928) states that only squamous epithelium of the skin and mucous membranes seems to be susceptible to the virus and that the basal layer of the epithelium shows little change from the normal. The early stages of the inclusion bodies appear peripheral to the basal layer. The cells in which they are present are enlarged, and the cytoplasm appears edematous. The material constituting the inclusion body appears first about the periphery of one or more vacuoles situated at the proximal pole of the cell. The inclusion body increases rapidly in size, in some cases becoming as large as the original cell. Nearer the surface, the altered cells become larger, and there is evidence of change in position of the inclusion bodies so that they come to lie more frequently at the distal pole of the cell. As the surface of the lesion is approached the cells show evidence of disintegration, and the inclusion body may become more

or less isolated in the cytoplasm. The increasing size of the inclusion body is accompanied by the ultimate destruction of the nucleus and the death of the cell. Extruded nuclear particles may appear in the cytoplasm. On the surface of the lesion there are great alterations of the cells from desiccation.

The presence of inclusion bodies in the affected cells of cutaneous eruptions of fowl pox was first discovered by Rivolta (1869). This discovery afforded a basis for morphological speculation as to the relationship of the inclusion bodies to the etiological agent of the disease. Bollinger (1873),
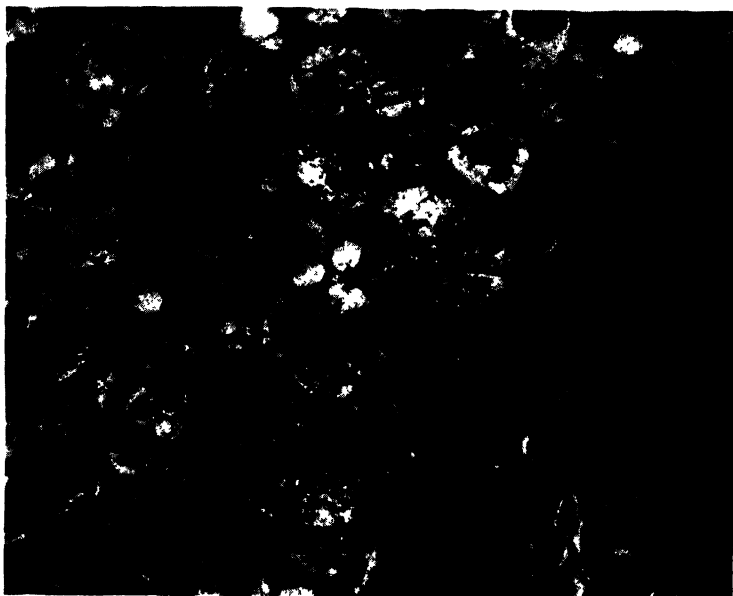


FIG. 25.1. Epithelial changes in fowl pox. ×460. (Biester, Iowa State College.)

through histological studies, differentiated the lesions of fowl pox and variola and classified fowl pox among the tumors as epithelioma contagiosum. This term unfortunately exists today but is an erroneous terminology, although the gross lesions of the disease, do have some macroscopic similarity to epitheliomata. Guarnieri (1892) demonstrated certain characteristic intracellular bodies in the lesions of variola differing from those of fowl pox. These studies of Bollinger and Guarnieri presented convincing pathological differentiations between the two diseases as distinct entities produced by different agents.

Borrel (1904) discovered in smear preparations fiom the cutaneous lesions of fowl pox myriads of minute coccus-like structures apparently small enough to pass through the pores of a Berkefeld filter. This discovery suggested that these bodies might be the specific etiological agent of the disease.

Burnet (1906) in a critical pathological study of fowl pox confirmed

Borrel's observations of the coccus-like structures in fresh preparations and showed that these bodies were derived from the same cell in which inclusion bodies could be demonstrated in stained preparations. Goodpasture (1928) reported that these bodies were about 0.25µ in diameter and may be seen easily under the microscope.

The conception of the causal agent of fowl pox at this period was that the specific cellular inclusion body or Bollinger body contained enormous numbers of elementary bodies or Borrel bodies which were the infective agent of the disease.

The experiments of Woodruff and Goodpasture (1929) on the nature and infectivity of inclusion bodies of fowl pox presented conclusive evidence as to the etiological agent of the disease. These authors discovered that a 1 per cent solution of trypsin in 0.2 per cent sodium bicarbonate would digest completely the cellular material of a fowl pox lesion in about 30 minutes, leaving the inclusion bodies free and separable from the tissue debris. The inclusion bodies were small, oval, occasionally bean-shaped or irregular, and varied from 2 or 3µ to perhaps 50µ. In their internal structure, they ranged from discrete granules to hyaline-like, homogeneous bodies. These inclusion bodies were interpreted as virus colonies containing enormous numbers of elementary bodies which were the causal agents of the disease. Inclusion bodies had an elastic, semipermeable membrane of lipo-proteid composition. The material which protected the inclusion body from tryptic digestion was a fatty element which was readily demonstrated by special fat stains. An albuminous component was also present in the inclusion. Inclusion bodies had a high density and quickly gravitated to the bottom of the container if suspended in physiological saline. In the presence of distilled water, inclusion bodies swelled with the formation of vacuoles and the elementary bodies exhibited Brownian movement. The addition of saline caused the inclusion bodies to shrink and assume again their initial hyaline, homogeneous appearance. Inclusion bodies could be washed free of trypsin by centrifugation and resuspending the bodies in saline. A 2 per cent solution of saline was better than physiological saline because of the reduction of surface tension which caused some of the inclusions to adhere to each other. Woodruff and Goodpasture concluded that a single inclusion body when washed in saline and inoculated into the skin of a susceptible chicken produced a typical fowl pox lesion containing the characteristic inclusions. The fluid in which the inclusion was finally suspended was innocuous.

In a later publication, Woodruff and Goodpasture (1930) showed that an inclusion body may contain as many as 20,000 elementary bodies each of which was capable of inciting the disease in susceptible fowl.

Histological studies by Woodruff and Goodpasture (1931) of chorio-allantoic membranes of embryonated chicken eggs infected with fowl pox

virus showed a marked susceptibility of ectodermal cells to infection, although entodermal cells could be infected. Inclusion bodies were usually less numerous and hyperplasia was less marked in entodermal cells than in ectodermal cells. A further indication that entoderm was less susceptible than ectoderm to fowl pox infection was seen in the fact that entodermal derivatives of the adult hen were rarely infected.

Brandly (1941) concluded from histological studies of infected chorioallantoic membranes of embryonated chicken eggs that "both fowl- and pigeon-pox viruses produced microscopic retrograde changes in all three germ layers of the chorio-allantois. Fowl-pox strains produced more severe alterations in the ectodermal and entodermal layers, while with the pigeon virus the reactions were more marked in the mesoderm (cellular infiltration and edema). The development of cytoplasmic inclusions (Bollinger bodies) produced by fowl strains appeared to be associated with necrosis of the limiting cellular layers. As a rule, these inclusions in the ectoderm and entoderm were larger and more readily demonstrated than those occurring in pigeon-virus lesions. Entodermal involvement by pigeon virus was invariably slight."

Groupé, Oskay, and Rake (1946) showed by electron microscopy that the elementary bodies of fowl pox closely resemble those of canary pox. These bodies have many characteristics in common with the elementary bodies of vaccinia. The pox bodies were approximately rectangular in shape, and occurred singly, in pairs, and in short chains. The bodies were most frequently attached to one another at the corners. The characteristically flattened corner frequently observed on many particles probably resulted from the separation of two such particles joined at their corners.

Groupé and Rake (1947) studied further the morphology of the elementary bodies of fowl pox and stated that "Although the elementary bodies of fowl pox are somewhat larger than those of vaccinia, the similarities between the two are striking. The particles of both appear to be approximately rectangular in shape and possess large central moundlike elevations. In addition, both seem to be coated with a sticky substance, and are frequently joined to one another at their corners. That classic examples of both avian and mammalian strains should so closely resemble one another morphologically adds still another link to the chain of evidence that has bound the viruses of the pox group together. The presence of forms suggesting unequal division of elementary bodies, together with the characteristics mentioned above, supports the suggestion of Green, Anderson, and Smadel (1942) that the pox viruses have morphological characteristics that approach those of the bacteria rather than those of the plant viruses." (Fig. 25.2.)

Burnet (1933a) observed well-marked cytoplasmic inclusion bodies in the epithelial cells of skin overlying lesions produced by Kikuth's canary pox

virus. There was a considerable resemblance of these inclusions to the Bollinger bodies of fowl pox. On the chorio-allantoic membrane of embryonated chicken eggs the virus produced massive inclusions in the proliferating ectoderm, but changes in the mesodermal and entodermal layers appeared to be purely secondary and no inclusion bodies were observed in the cells.

Durant and McDougle (1938) reported the findings of their histological studies of canary pox as follows: "Microscopic studies of the stained sections



FIG. 25.2. Fowl pox virus shadowed with gold. Electron-micrograph. 2.3 x 12,400×. (Groupé and Rake, 1947.)

from canary-pox lesions in canaries, chickens and turkeys showed distinct differential characteristics. In the sections from canaries numerous typical virus inclusion bodies are present with very few if any polymorphonuclear eosinophils with rods. In the section from chicken lesions there are no virus inclusion bodies evident and only a few polymorphonuclear eosinophils with rods. In the case of turkeys the sections showed a marked infiltration of polymorphonuclear eosinophils with rods with small bodies resembling virus bodies which are usually seen in the canary pox or fowl pox lesions. These are observed only in the deeper tissues . . . . "

Coulston and Manwell (1941) observed typical cellular inclusions in canary pox.

**Diagnosis.** The presence of cutaneous lesions typical of fowl pox infection in chickens usually warrants a diagnosis of the disease. A diagnosis is not so readily made when mouth cankers or coryza-like lesions and symptoms are seen. Several diagnostic tests may be employed, viz., infectivity tests, protection tests, microscopic examination of lesions, and serological tests as described by Brandly and Dunlap (1938).

With infectivity tests it may be necessary to utilize heterologous as well as homologous hosts for an accurate diagnosis of the type of pox virus present in the bird. The presence of fowl pox virus in suspected lesion material may be demonstrated readily by the application of an emulsion of the material to the skin of susceptible chickens by scarification of the comb or by the "stick" and "feather follicle" methods described later. If the virus is present in the inoculum the chicken will develop typical cutaneous lesions of the disease in from 5 to 7 days. In cases where the suspected lesion material or the induced lesions are not typical, microscopic examinations should be made of scrapings from the base of the lesions for detection of elementary or Borrel bodies.

Protection or immunity tests with fowl pox immune and susceptible chickens may be used simultaneously with infectivity tests and microscopic tests. Exposure of the immune and susceptible birds to suspected lesion material containing fowl pox virus by the methods described above will result in refractivity in the immune bird and the development of typical cutaneous lesions in the susceptible bird. The original fowl pox suspected birds which recover from the infection may be tested for immunity to a known fowl pox virus.

Microscopic examinations of smears and histological preparations of suspected lesion material are of diagnostic value. Smears of the lesions may be prepared according to the method of Goodpasture (1928) : "If the surface of such a lesion be slightly scraped and the scrapings moistened with water and pressed under a coverglass, the virus bodies may be easily recognized under the microscope. When they are pressed out thin beneath the cover-glass they are seen to be composed of myriads of minute bodies which seem to be agglutinated by a viscous material between them. In smears stained with Loeffler's flagella stain, or better with carbol-anilin-fuchsin for 1 minute, after mordanting an equal period with ¼ per cent potassium permanganate (Goodpasture), they are readily demonstrable. They are round or slightly oval, sometimes arranged in short chains or in diplococcal and biscuit forms. They are colored a distinct pink by the latter method, and are Gram-negative. These bodies may be studied equally well in dark-field preparations. They measure about $0.25\mu$ in diameter, and have the appearance generally of a minute, nonmotile microorganism."

Brandly and Dunlap (1938) utilized the method of Morosow (1926) for preparing and staining direct smears from suspected lesion material. These authors have successfully used this method of examination to facilitate rapid diagnosis of suspicious field cases.

Stained histological preparations should reveal the presence of Bollinger bodies and the typical microscopic lesions observed with fowl pox infection.

Serological tests of the subject virus with known fowl pox antiserum may be conducted with living hosts and also with embryonated eggs as described by Burnet (1936b) and Burnet and Lush (1936) for neutralization tests. Serum with high neutralization and virucidal properties may be produced by hyperimmunization of chickens. Burnet and Lush (1936) produced fowl pox antiserum by inoculation of susceptible birds on the scarified comb and intramuscular injection of an emulsion of fowl pox infected chorio-allantoic membrane material. A course of three more intramuscular injections of infected membranes was begun three weeks after the initial inoculation. Blood was collected from the birds 10 days after the last inoculation.

Neutralization tests utilizing embryonated chicken eggs would necessitate the recovery of the virus in a bacteria-free inoculum. Technics for this procedure and propagation of the virus on the chorio-allantoic membrane are discussed under the section *Cultivation of avian pox viruses.* Isolation and propagation of the virus in eggs with the appearance of typical lesions of fowl pox infection on the chorio-allantoic membrane would offer presumptive evidence of the identity of the virus. Identification of the virus requires neutralization tests using the embryo technic, protection or immunity tests, or serological tests.

Dalling, Mason, and Gordon (1929) were successful in producing fowl pox antiserum by immunizing susceptible chickens through application of the virus to the scarified comb. After "takes" were no longer produced by subsequent application of the virus to the comb, a course of four to six increasing doses of active virus was injected intramuscularly. The authors reported that the protective value of the antiserum could be estimated by two methods—scarification and intravenous injection. With the scarification method, equal amounts of the test-dilution virus and antiserum were admixed, incubated at room temperature for 1 hour, and then applied to the scarified comb. It was possible to test as many as twelve samples on the comb of one bird. With the intravenous method, varying amounts of antiserum were injected intramuscularly, and on the following day each bird received the test-dilution of virus intravenously.

Ledingham (1931) demonstrated agglutination of the elementary bodies of fowl pox virus with sera in dilutions as high as 1:160 from recovered and hyperimmunized chickens. Agglutination did not occur with normal sera. Quantitative experiments (Ledingham, 1932) showed that the agglutinin

response was slow after inoculation with the virus. In one instance, agglutinins did not appear until the seventeenth day, and sera from recovered chickens showed titers varying from 1:10 to 1:80 at periods varying from four months to one month. At later periods (up to seven months) the sera from these same birds were devoid of agglutinins. Sera from hyperimmunized birds showed titers as high as 1:300.

**Cultivation of avian pox viruses.** Fowl pox virus like other viruses can be cultivated only in the presence of living cells of susceptible hosts. These requisites may be supplied by the living host, avian embryos, and to a lesser degree by tissue culture.

Cultivation of fowl pox virus in the living host may be accomplished by application of the virus to the scarified comb of a susceptible chicken and collection of the scabs. Brandly and Bushnell (1932) reported that lesion material harvested the tenth and eleventh days after inoculation was the most virulent and produced the most extensive cutaneous lesions. The scabs should be desiccated, powdered, and stored under refrigeration until used for the next passage. Pigeon pox virus may be propagated according to Graham and Brandly (1940) by the application of a freshly prepared 1 per cent aqueous solution of powdered pigeon pox skin lesion material to a defeathered area of the ventral surface of the breast of pigeons. Inoculated pigeons were killed when in a moribund condition, usually about the sixteenth day, and the affected skin and scab removed. This material was desiccated, cut into small pieces, ground to a fine powder, and refrigerated until used for the next passage.

The technic of avian embryo culture of viruses requires bacteria-free inoculum. Woodruff and Goodpasture (1931) successfully cultivated the virus of fowl pox on the chorio-allantoic membrane of embryonated chicken eggs, and the authors recorded three methods for the collection of bacteria-free fowl pox virus. In the first method the feathers are plucked from the heads of one- to two-week-old chicks and the virus inoculated at three points about 1 cm. apart. On the sixth or seventh day after inoculation the chick is sacrificed as at a later date the pox nodules are likely to be contaminated with pyogenic bacteria. The head of the chick is bathed with 95 per cent alcohol and allowed to dry. A sterile scalpel is used to cut off the pox infected nodules at a point deep enough to obtain the infected core of most of the nodules. The infected core is then forced out of the follicle from the cut surface. The cores are washed twice with Tyrode's solution and stored at 4° C. in the same solution. Bacteriological sterility tests of the collected material are made with glucose yeast broth. If the material is bacteriologically sterile it is ground with Tyrode's solution and used as a source of virus inoculum.

In the second method, 7- to 10-day-old lesions of fowl pox are collected

and subjected to digestion in 1 per cent trypsin solution to free the inclusion bodies from the tissues. The inclusion bodies are then washed several times with sterile saline and a single inclusion is picked up with the pipette of a Chambers microdissection apparatus and deposited on the chorio-allantoic membrane of the embryo.

In the third method, the infected pox lesions are digested with trypsin to liberate the inclusions which are then treated with a 1 per cent solution of potassium hydroxide for 24 hours. The authors state that treatment with potassium hydroxide solution of the pox lesions without previous digestion with trypsin fails to render the material free of certain molds and bacteria.

Beaudette and Hudson (1938) reported the successful modification of Woodruff and Goodpasture's first method with fowl pox virus. These authors inoculated the feather follicles on the leg of a half-grown chicken with fowl pox virus and sacrificed the chicken when the lesions developed. The feathers were removed from the leg and the skin singed carefully. The skin was raised from the leg and scrapings were made from the deeper portions of the skin with a sterile scalpel. The small amount of material thus obtained was inoculated directly onto the chorio-allantoic membrane of embryonated eggs. Control tests for bacteriological sterility were made on agar culture media. This procedure was used by Beaudette and Hudson (1941) for the collection of bacteria-free turkey virus. Beaudette and Hudson (1938) found that collection of pigeon pox virus from deep scrapings of infected feather follicles was of no value, as heavy contamination of the harvested material was always experienced. They resorted to filtration of the supernatant fluid from a ground emulsion of infected skin from chickens infected with the pigeon pox virus. Berkefeld V filtrates of the fluid were contaminated as demonstrated by sterility tests, but it was found that the membrane of one egg at the time of harvest had only a slight amount of bacterial contamination and another membrane was bacteria-free. The bacteria-free membrane was used to initiate further passage of the virus. It appeared in this case that the few bacteria passing the filter were destroyed by the bactericidal substances in the egg.

The Bierbaum and Gaede (1935) method of intracerebral inoculation passage through pigeons and/or chickens of pigeon pox lesion material for obtaining bacteria-free virus inoculum has been utilized successfully by Brandly and Dunlap (1938), and Brandly (1941). This method consists of introducing the material intracerebrally, and after 8 to 12 days incubation the bird is sacrificed and the brain removed aseptically. Brandly (1941) reported that in two instances a pure pox virus was obtained from the first passage and rarely were three or more intracerebral passages necessary to free the virus from bacteria.

The utilization of avian embryos for virus culture by Woodruff and

Goodpasture (1931) is of relatively recent date, but since that time a mass of published data has appeared on this technic of cultivation of viruses. Avian embryo culture offers an economical and convenient means for the pursuit of many fundamental virus investigations as well as the source of materials rich in virus content for the production of viral immunization agents free of bacterial contamination. The source of eggs should be vigorous, disease-free breeding stock, and only strong, well-developed embryos should be selected for virus cultivation. Consideration must be given to the species of embryos, as certain viruses fail to propagate in embryonated eggs of all domestic fowl. The temperature of incubation and the age of the embryos also influence the action of the virus, as well as the route of inoculation.

There are two general methods, with modifications, for preparing eggs for inoculation of the chorio-allantoic membrane with virus. The Brandly (1935, 1936, 1941) method consists of introducing the needle of the syringe through a hole in the shell over the air cell and inserting the needle between the inner shell membrane and the chorio-allantoic membrane where the inoculum is deposited. The hole in the shell is sealed with discs of colorless cellophane applied with library paste, or cellulose tape (transparent) may be substituted for the cellophane. The eggs are then returned to the incubator for further incubation and observation.

The Burnet (1936a) method, with modifications, consists of the production of an artificial air cell on the side of the egg. A hole is drilled through the normal air cell and another hole is drilled through the shell on the side of the egg, or a triangle, square, or disc may be cut out of the shell on the side of the egg. With the egg in the horizontal position to its long axis, the shell membrane is punctured and slight negative pressure is applied to the hole over the normal air cell. The chorio-allantoic membrane will drop from the side of the shell causing the egg contents to occupy the normal air cell. The inoculum is deposited on the chorio-allantoic membrane of the artificial air cell and the opening in the shell is closed with melted paraffin, cellulose tape, cellophane applied with library paste, or a cover glass held in position on a ring of paraffin.

The use of transparent materials for closing the opening in the shell in both methods allows direct observation of the development of lesions on the chorio-allantoic membrane during the subsequent incubation period.

The amount of inoculum to be employed for avian embryo cultivation of fowl pox virus depends upon the particular problem under consideration. Inoculum of 0.05 cc. or more has been reported by various investigators.

Woodruff and Goodpasture (1931) used 10- to 15-day-old embryos for their investigations of fowl pox virus. They were, however, successful in cultivating the virus on the chorio-allantoic membrane of 6-day-old, and,

in one instance, 4-day-old embryos. Slight abrasion of the skin of the embryos as employed in one of their methods of cultivation resulted in successful growth of the virus, but the trauma resulting from this method was so great and the mortality so high that this method was abandoned.

Burnet (1936a) reported that for the study of inclusions 10-day-old embryos are best suited, while for virus production 11- or 12-day-old embryos are preferred. Brandly (1936, 1937, 1941) used 10- to 14-day-old embryos, but 12-day-old embryos were preferred for critical observations of the action of the virus.

Brandly (1937) studied the susceptibility of duck, guinea fowl, and turkey eggs to fowl pox virus as compared to chicken eggs. The eggs were of various ages from 10 to 18 days with the control chicken eggs 12 days old. Infection was obtained in all of the species of eggs employed but a ten-to thirty-fold greater end-point concentration of the virus was necessary to initiate infection in the duck eggs. This was interpreted as a lower degree of susceptibility of the duck-egg membranes to infection with fowl pox virus. An apparent increase in the resistance of duck eggs to fowl pox infection was noted after the fifteenth day of incubation. Infection was not noted in the duck eggs inoculated on the eighteenth day while slightly more than half of the eggs inoculated on the sixteenth day showed evidences of infection. Turkey eggs of the same age as the duck eggs and inoculated simultaneously with the same virus preparation showed infection of 80 per cent of the eggs. With chicken eggs it was found in some instances that infection was obtained in 10- and 12-day-old eggs with virus concentrations approximately ten to thirty times smaller than was required to infect 14-day-old eggs. A tendency to metastasis of pock lesions was noted on the membrane of the 10- and 12-day-old eggs when dilute suspensions of virus were used. Large confluent lesions confined to the large pole of the egg were found in about equal numbers of the eggs of the different ages which received concentrated virus suspensions. Differences in the survival time of the embryos in eggs of various ages that developed pox infection did not appear consistent or significant. The infective concentration of fowl pox virus was not materially influenced by adsorption when powdered Pyrex glass and quartz sand were used as abrasives for grinding infected egg membranes for preparation of inoculum. Repeated egg passages through twenty successive series of eggs did not change the virus insofar as the appearance of skin lesions in vaccinated birds was concerned. It was recommended that all harvested material be rapidly dehydrated at a low temperature if the material was to be held an appreciable length of time.

Brandly and Dunlap (1939) reported that passage of one strain of fowl pox virus through sixty-eight series of eggs did not influence the virulence of the virus for chickens or for the chorio-allantoic membrane of the embryo-

nated egg. The membranes of 12-day-old eggs were found to be more susceptible to infection with fowl and pigeon pox virus than was the skin of chickens six to twelve weeks old.

Brandly (1941) conducted extensive experiments on the propagation of fowl and pigeon pox viruses in embryonated chicken eggs and the utilization of these viruses in immunization studies. Inoculated 12-day-old embryos were incubated for an additional 4 to 5 days and the chorio-allantoic membranes were collected for further studies. Gross lesions of the chorio-allantoic membrane were visible as early as 48 hours after inoculation with the viruses. The pigeon pox viruses had the tendency to localize over the area inoculated. The fowl pox viruses tended to metastasize rapidly. According to Brandly "The nature of the lesions induced in developing chicken eggs by fowl-pox and pigeon-pox viruses differed considerably among the strains studied. Grossly, the pigeon-virus lesions were typically pale yellow to white, with a nacre or pearly tint, whereas the fowl-virus infected membranes were usually reddish gray and quite heavily congested. Individual pocks, . . . . were globular in form in the case of pigeon virus, compared with the somewhat thinner and relatively flat fowl pocks." (Fig. 25.3.)

Thorning, Graham, and Levine[*] (1943a) presented evidence of the presence of fowl pox virus in the embryo proper and yolk as well as in the chorio-allantoic membrane. The greatest concentration of the virus was in the chorio-allantoic membrane, a lower concentration in the yolk, and a still lower concentration in the embryo proper. Thorning, Graham, and Levine (1943b) in a later contribution stated that "Available evidence does not indicate that multiplication of fowl pox virus occurs in various parts of the embryo exclusive of the chorioallantois, and until further evidence is presented, it appears that the virus content, as well as its immunogenic activity, is related to the virus content of the chorioallantois."

In Beaudette and Hudson's (1938) report on the cultivation of pigeon pox virus on the chorio-allantoic membrane of chicken eggs, it is stated that the lesions produced by the virus did not differ in appearance from those of fowl pox virus. The most extensive involvement was seen when the inoculum was rich in virus and the eggs were incubated more than 5 days following inoculation. Membranes removed from eggs near the hatching date contained little or no virus. One trial with 8-day-old pigeon eggs showed by inoculation of chickens with the harvested membrane that the virus was active, but there was no evidence of the growth of the virus on the membrane since lesions were not produced. Fowl pox virus was apparently lethal to 12-day-old Muscovy duck embryos whereas it has but little effect or lethal action on chicken embryos. The Muscovy duck embryos were red, but the membranes were not thickened and showed no lesions. Although the Muscovy duck embryo membranes showed no evidence of infection, they were rich

in virus as evidenced by a "take" by the "feather follicle" method when inoculated into chickens and as evidenced by a heavy infection on the inoculated chorio-allantoic membrane of 10-day-old chicken embryos. Chicken-embryo-membrane propagated fowl pox virus produced extensive
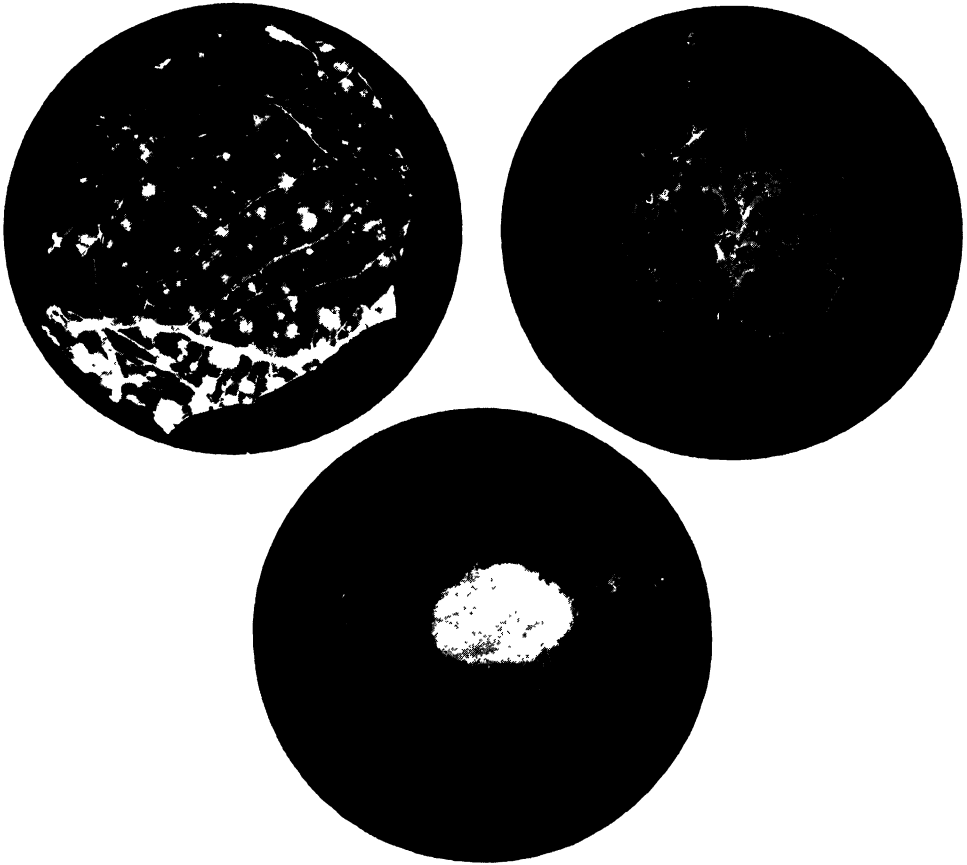


FIG. 25.3. Fowl and pigeon pox lesions on chorio-allantoic membrane. A—metastatic fowl pox lesions. B—diffuse fowl pox lesions. C—focal pigeon pox lesion. (Brandly, Ill. Agr. Exper. Sta., Bul. 478.)

lesions on the chorio-allantoic membrane of 12-, 13-, and 14-day-old Pekin duck embryos.

The first report of the successful cultivation of turkey pox virus in embryonated chicken eggs was made by Beaudette and Hudson (1941). The virus was obtained in the form of a dried scab removed from a wild turkey. The scab was kept in an electric refrigerator for nearly eight years when it was emulsified and passed through a Berkefeld V filter. The filtrate was used

to initiate infection in chicken eggs but apparently did not contain virus, as typical evidence of pox infection was not seen on the membranes, and transfer of emulsions of these membranes in other eggs gave negative results. A portion of the sediment of the emulsion was used to vaccinate a chick by the "feather follicle" method. Deep scrapings of the skin near the few infected follicles was passaged through eggs which showed heavy infection. Since the membranes were contaminated, Berkefeld filtrates were prepared and further eggs inoculated. These eggs showed definite evidence of infection. There were no detectable differences in the macroscopic appearance of the lesions produced by this virus and that of fowl pox, but the turkey pox virus was less lethal for the embryo than the fowl pox virus.

Burnet (1933a) reported that the canary pox virus of Kikuth can be cultivated on the chorio-allantoic membrane of embryonated chicken eggs and is capable of producing a massive lesion or plaque of grayish-yellow thickening of the membrane. The lesion generally resembles that produced by typical strains of fowl pox virus but it is recognizably different.

Brandly and Dunlap (1939) employed several methods of tissue-culture propagation of pigeon pox and fowl pox viruses. A minced chicken-embryo-Tyrode solution medium gave the most satisfactory results. Continuous cultivation of the viruses in this medium was not found difficult, and concentrations of the virus were suitable for immunization procedures, but this method was not as satisfactory as propagation of the viruses in embryonated chicken eggs.

**Effects of certain physical and chemical agents on the virus.** With the exception of a few instances in which purified preparations of viruses have been used, all experiments to determine the virucidal effect of certain physical and chemical agents have been conducted with preparations containing tissue elements and extraneous protein material. Investigations of the effect of these agents on the fowl pox virus have been conducted with impure preparations of the virus, and the data may be interpreted only on a relative basis. Lack of uniformity of quantitative standardization of the virus concentration also hinders assessment of the effect of the agents.

One of the characteristic properties of fowl pox virus is its resistance to desiccation. The virus is well adapted to survival for long periods when exposed to climatic conditions such as dryness, moisture, and light, especially during cold seasons. Moist heat destroys the virus fairly readily. The resistance of the virus to desiccation is an important factor in its persistence under natural conditions.

According to Burnet (1906) aqueous or physiological saline suspensions of the virus in sealed glass ampoules were inactivated within 8 minutes at 60° C., but infected nodules and scabs were still active after 1½ hours, and finely powdered virus material did not survive for more than ½ hour. A

fragment of nodule imbedded in agar was active after 14 days at 38° C. Aqueous suspensions of virus were active after 60 days at 6° C., 30 days at 22° C., 6 days at 25° C., and 3 days but not 8 days at 37° C. A finely powdered suspension of virus added to an equal volume of glycerin was active after 120 days at 30° C. The glycerin had a bacteriostatic effect as very few organisms could be detected in the suspension after 60 days. A finely powdered preparation of the virus which was spread upon glass slides and slowly dried was found to be active after fifteen months. Virus dried in vacuum over $H_2SO_4$ was active after twelve months.

According to von Reischauer (1906) the virus resists dry heat for 15 to 30 minutes at 80° C. and moist heat for 5 minutes at 100° C. The virus is killed in 5 minutes by 1 per cent potassium hydroxide, 1 per cent acetic acid, and bichloride of mercury 1:1,000. Loewenthal (1906) exposed the virus to radium and found it active after $5\frac{1}{2}$ hours. An emulsion of scabs were active after $1\frac{1}{3}$ hours in 1 per cent phenol but not in 2 and $2\frac{1}{2}$ per cent phenol according to Marx and Sticker (1903). Graham and Barger (1936) found that 1 per cent aqueous suspension of fowl pox virus on sterile cotton squares, on the feet and down of day-old chicks, upon being subjected to routine incubator fumigation survived 30 minutes, often 45 and 50 minutes, but was consistently noninfective after 90 minutes. The infectivity of the virus suspension fumigated for 30 and 45 minutes was not appreciably altered as demonstrated by infectivity tests with susceptible chicks.

Graham and Brandly (1940) reported that 1 per cent suspensions of virus containing 0.025 to 0.5 per cent formalin, 0.5 per cent phenol, 2 per cent saponin, and 0.5 per cent tricresol were completely inactivated when stored for 48 hours at icebox temperatures. Coulston and Manwell (1941) found that canary pox virus in the dried form was virulent for seven months but not for eleven months. Beaudette (1941) reported that a dry scab removed from a wild turkey and stored in an electric refrigerator contained active virus approximately eight years later.

McCulloch (1945) enumerates several factors to be considered for an assessment of the virucidal properties of physical and chemical agents. The author reported that 95 per cent and 75 per cent solutions of ethyl alcohol inactivated the virus in less than 10 minutes, 50 per cent ethyl alcohol inactivated the virus in 30 minutes but not in 10 minutes, and 25 per cent ethyl alcohol was without effect on the virus. These tests were made at 20° C. with 1,000 infective doses, and the broth in which the finely suspended virus material was tested was at pH 7. The virus was found to be inactivated by 50 parts of available chlorine per million when the material was suspended in F.D.A.[1] broth. The virus was able to withstand 20 minutes exposure to 3 per cent formaldehyde solution at 20° C., although the incubation period in

[1] Food and Drug Administration.

chickens inoculated with the treated virus was prolonged. A commercial solution of hexylresorcinol (1:1,000) inactivated the virus when diluted 1:4 but not when diluted 1:8. Tincture of iodine diluted 1:400 inactivated the virus, but a 1:800 dilution was without effect. A 1 per cent aqueous solution of mercurochromé was without effect while a 2 per cent solution inactivated the virus at 20° C. at pH 7. The virus was able to withstand 3 per cent phenol for 10 minutes at 20° C. but not for 30 minutes. A 1:500 dilution of sodium hydroxide at 20° C. inactivated the virus in 10 minutes, but a 1:600 dilution was without effect. When the virus was suspended in F.D.A. broth pH 7, at 20° C., 1:1,000 crystal violet inactivated the virus in 10 minutes, 1:50 acriflavine in 10 minutes but not 5 minutes, while 1:100 acid fuchsin failed to inactivate the virus in 30 minutes. In each inoculum there were approximately 1,000 infective doses. A 1:400 dilution of liquor cresolis at 20° C. inactivated the virus in 10 minutes but not in 5 minutes, while at 20° C. a dilution of 1:5,000 was effective as the same dilution at 40° C.

A suspension of 100,000 infective doses of the virus in F.D.A. broth was inactivated in approximately 5 minutes at 60° C., in 15 to 20 minutes at 55° C., and resisted for longer than 1 hour at 50° C.

Graham, Brandly, and Levine (1939) reported that irradiation of aqueous suspensions of fowl pox virus with hard X-rays in dosages up to 888 r units had no effect on the virus. The virus was inactivated by ultra-violet light from a mercury vapor lamp in 2 hours at 20 cm. and attenuated when exposed for 15 to 90 minutes. The addition of methylene blue in a concentration of 1:50,000 reduced the time necessary for inactivation to 5 minutes, while 2½ minutes irradiation markedly attenuated the virus.

Robbins (1944) reported that neither penicillin nor patulin were effective against the virus as shown by infectivity tests on the chorio-allantoic membrane of embryonated chicken eggs.

**Epizoology.** Fowl pox is a widespread disease and is prevalent wherever poultry is raised. The incubation of the spontaneous disease varies from 4 to 6 days, according to Goodpasture (1928), and from 6 to 14 days, according to Delaplane (1943). While the disease may make its appearance at any time, the greatest incidence of infection is during the fall and winter months. A few outbreaks may occur during the spring, and the disease is uncommon during the summer. Under natural conditions, the disease usually makes its appearance in young stock at about the time they are housed in laying quarters. Following this the disease may reach serious proportions. During the fall and early winter, the cutaneous form of the disease predominates in most outbreaks, while during the winter months the diphtheritic form is usually most common. The epizoological picture of fowl pox is similar to that of other contagious diseases in that variations of the virulence of the disease may be observed. In some outbreaks the majority of the flock may be

affected within a short time, while in other flocks the disease may spread more slowly. If nothing is done to control an outbreak, it may persist in a flock throughout the winter. The course of the uncomplicated disease is usually about three to four weeks, but if complications are present the duration may be considerably longer. Recovered birds are immune to further infection.

Chickens affected with the cutaneous form of the disease are more likely subjects for recovery than chickens affected with the diphtheritic form of the disease particularly when the lesions involve the respiratory tract, eyelids, and nasal sinuses. A flock of chickens in good physical condition usually warrants a favorable prognosis, while the prognosis is unfavorable if the flock is affected with other infectious or parasitic diseases or is subject to poor nutrition and management.

The mortality rate is variable. In some flocks it may be negligible while in others it may be high. In laying flocks the egg production will be temporarily retarded. This is associated with an increasing number of emaciated chickens. The mortality in laying flocks may assume serious proportions.

Individual symptoms of fowl pox infection may be manifested in one of three forms or a combination of these forms depending upon the virulence and pathogenicity of the strain of virus involved: (1) localization of typical cutaneous pox lesions on the comb, wattles, and face region (Fig. 25.4); (2) localization of the infection in the mouth region with the appearance of typical diphtheritic lesions; and (3) localization of the infection in the nasal chambers with accompanying coryza-like symptoms. While pox lesions are not commonly observed on the feet, legs, and body of chickens, these lesions may be observed if chickens are reared on wire floors. Other than the typical cutaneous and diphtheritic lesions produced by the disease there are no constant or characteristic lesions to be found at post-mortem examination of affected chickens.

Chickens of all ages, sexes, and breeds, unless previously exposed, are equally susceptible to the virus by inoculation. Under natural conditions, there seems to be possible breed differences in susceptibility. Cary (1906) reported that chickens with large combs seem to be more susceptible to infection than chickens with small combs. Johnson (1927) has reported that Leghorns appear to be more susceptible to natural infection than are Barred Plymouth Rocks because of the large comb area.

The disease is not commonly seen in young chickens, although Beaudette (1929) and Johnson (1938) have observed outbreaks in battery brooded chicks. In the latter report, the chicks were six weeks old, and the lesions in practically all of the cases were found on the feet and legs. The absence of lesions on the combs and wattles were attributed to the lack of development of these organs at this age.

Fowl pox virus is unable to penetrate intact epithelium. Application of

the virus to scarified epithelium or mucous membranes of the mouth, and to defeathered feather follicles will result in establishment of the infection. Doyle and Minett (1927) were unable to induce infection by the daily application of virus to intact epithelium of the combs of chickens. The application to the leg of pledgets of cotton soaked in virus likewise failed to infect chickens. They were also unable to produce infection in fowls which were fed virus in capsules. Injection of virus subcutaneously, intramuscularly, intraperitoneally, and instillation in the conjunctival sac resulted in infec-



Fig. 25.4. Severe case of fowl pox. (Brunett. Cornell Vet.)

tion. Intravenous injection of the virus produced the disease in three forms: (1) generalized infection with skin or mouth lesions; (2) slowly progressive emaciation without lesions; and (3) immunity without lesions or loss of condition. In transmission experiments, these authors showed that when susceptible chickens were placed in association with infected chickens, the susceptible chickens contracted the infection if picking was not prevented. In the absence of picking, and the exposure of susceptible chickens to an environment previously contaminated by infected chickens, or exposure in experimentally infected cages, the disease was not transmitted to susceptible chickens. The authors concluded that infection of the skin resulted from injuries sustained in picking, and that mouth lesions were the result of

injuries produced by eating of grit. The intimate cohabitation of chickens in most flocks enhances the possibility of spread of the infection since most chickens in a flock present injured skin surfaces so that there is no lack of suitable portals of entry of the virus.

The possibility of "carrier" chickens serving as foci of fowl pox infection has been the subject of considerable speculation and research work. According to Doyle and Minett (1927) and Doyle (1930) several investigators have indicated that pigeon pox virus may localize in certain internal organs, and may persist for a considerable length of time in these organs in recovered pigeons. Doyle (1930) stated it is possible that the frequently repeated statement that fowl pox virus acts in a similar manner is based on this work. Doyle and Minett (1927) found that fowl pox virus could be demonstrated in variable quantities in the blood of chickens throughout the course of the disease following intravenous injection of the virus or application of the virus to scarified areas of the comb and in the mouth. They infected chickens on the comb and in the mouth, killed them at intervals varying from 7 to 44 days after inoculation, and attempted to establish the presence of the virus in the various organs by inoculation of susceptible chickens. In no instance was the virus demonstrable in the internal organs of recovered chickens or on the comb after complete disappearance of lesions.

According to Burnet (1906) if feathers are plucked or the skin is scarified after intravenous injection of the virus, specific lesions will develop at the site of the cutaneous injury. The author summarized that the requirements for the development of eruptive lesions are the virus in the circulation and a susceptible point for the lesion.

Beaudette (1941) stated that in a small percentage of birds (usually less than 5 per cent) secondary lesions may appear on the head as a result of the virus having been transported to the area through the circulatory system.

That fowl pox virus may be transmitted by intermediary carriers has been reported by several investigators. Cary (1906) stated that "Mosquitoes—and other insects—may sometimes be the real carriers of the real virus." Kligler, Muckenfuss, and Rivers (1929) have shown that two species of mosquitoes, *Culex pipiens* and *Aedes aegypti* are capable of transmitting the disease from infected to susceptible chickens, as lesions developed in from 5 to 10 days after the infected mosquito was allowed to feed on a susceptible chicken. The mosquitoes were considered to be mechanical carriers of the virus as they were capable of transmission of the disease immediately after feeding on infected chickens. In one case the mosquitoes remained infectious for at least 14 days. Healthy chickens placed in a mosquito-proofed cage with an infected chicken and recently hatched *Culex pipiens* mosquitoes contracted the disease whereas under the same conditions, except that the mosquitoes were excluded, transmission of the disease did not take place.

Kligler· and Ashner (1929) studied the transmission of fowl pox by mosquitoes, *Culex pipiens* and *Aedes aegypti,* and showed that the same mosquito may produce a number of consecutive infections over a period of at least 16 days. Infected mosquitoes were capable of transmitting the infection despite intermediate feeding on guinea pigs. These investigators found that the virus appeared to be localized on the proboscis of the mosquitoes which may remain infective for 16 to 19 days. Only rarely could the disease be transmitted by inoculation of other parts of the mosquito's body. The virus behaved in the same manner on infected pins as on the proboscis. Blanc and Caminopetros (1930) showed that infected *Culex pipiens* could transmit the infection for at least 58 days following an infective meal and from pigeon to pigeon for at least 38 days. Matheson, Brunett, and Brody (1932) showed that the virus could be transmitted by *Aedes vexans* for at least 27 days following an infective meal.

Stuppy (1932) presented evidence that *Culex pipiens* and *Stegomyia fasciata* were capable of transmitting fowl pox infection for at least 39 days after feeding on an infected chicken. The incubation of the mosquito-transmitted infection was from 6 to 8 days. The experimental infection produced by the infected mosquito was exactly the same as the natural infection and was sufficient to produce immunity to further attacks. It was believed that the transfer of the infection from an affected chicken to a susceptible chicken was not simply mechanical since the infectiousness of the mosquitoes was undiminished after 39 days. It was believed probable that the mosquitoes remained infective for their entire lives. Experiments by Stuppy indicated that the virus was present in the body of the mosquitoes.

The incubation period of Kikuth's (1932) canary pox is about 4 days, the canaries dying from 7 to 12 days after infection. Durant and McDougle (1938) observed that canary pox apparently occurs in cycles of about 21 days in young canaries with a mortality of about 100 per cent of all birds affected. Death occurred quite regularly from the tenth to the fourteenth day after exposure. Recovered canaries were refractory to subsequent exposure to a virulent virus. According to Coulston and Manwell (1941) canary pox was uniformly fatal to canaries after a week or 10 days when the lesions were localized other than on the toes and legs. In the latter form the disease was chronic, but it also killed the canary after a period of some weeks or months.

**Prevention and control.** Efforts to develop an effective, dependable prophylactic agent for immunization against pox in domestic fowl have long been the subject for studies of the disease. Graham and Brandly (1940) present an extensive review of the literature on immunization. Prior to 1902 when the etiological agent of the disease was determined to be a filtrable virus, the results of the investigations had not been successful. Since that time effective prophylactic agents have been developed. Much of the early

work was conducted with virus which had been attenuated, and in some cases inactivated, by physical or chemical means, or virus modified by passage through heterologous hosts. The immunogenic properties of these preparations were subject to wide variations. Recognition of the ineffectiveness of these vaccines prompted the investigators to explore the possibilities of the utilization of virulent fowl pox virus obtained from cutaneous lesions of the disease. De Blieck and van Heelsbergen (1923) were probably the first to use on a large scale the fully virulent fowl pox virus for cutaneous immunization against fowl pox. Results indicating that vaccination of chickens with fully virulent fowl pox virus is an effective immunizing agent have been reported by many investigators who have also emphasized that certain potential post-vaccination hazards may accompany this method of immunization. A review of the reasons for failures in immunization against pox has been presented by Beaudette (1941).

Two types of vaccines are available for immunization of domestic fowl against pox: fowl pox vaccine and pigeon pox vaccine. The success of an immunization program with these vaccines depends upon their utilization only under the conditions where indicated, and upon the potency and purity of the vaccines and their application.

*Fowl pox vaccine.* Fowl pox vaccines are of two types: "chicken origin" and "chick-embryo origin" or "egg-propagated," depending upon the method of preparation.

The "chicken origin" vaccine is prepared by propagation of the virus on the scarified combs of chickens. The scabs or lesions are collected, desiccated, ground to a fine powder, distributed in suitable containers, and stored under refrigeration. Brandly and Bushnell (1932) reported that scabs removed the tenth and eleventh days after inoculation were the most virulent, whereas Johnson (1934) reported removal of scabs two to four weeks after inoculation. Some investigators report that a six months storage period should be considered the maximum limit for maintenance of potency of the virus. When the virus is to be used it is suspended in a suitable diluent such as sterile distilled water, physiological saline, or 50 per cent glycerin. Beaudette (1929) reported that 1 gram of virus suspended in 40 cc. of diluent was a suitable concentration of the virus for effective immunization. Graham and Brandly (1940) and Barger and Card (1943) report the use of a 1 per cent aqueous suspension of the virus. Barger and Card (1943) suggest the addition of 10 per cent glycerin to increase the adherence of the vaccine to the skin. A review of the literature reveals wide variations of concentrations of the vaccine preparations.

The "chick-embryo origin" or "egg-propagated" vaccine is prepared by propagation of the virus on the chorio-allantoic membrane of embryonated chicken eggs. The infected membranes are collected, desiccated. ground to

a fine powder, distributed in suitable containers, and stored under refrigeration. The best type of containers are glass vials or ampoules in which the virus can be hermetically sealed in vacuo. Storage under these conditions is conducive to maximum retention of potency of the virus. A suspension of 40 mgs. of virus in 2 cc. of diluent is satisfactory for the vaccination of 100 chickens according to Beaudette (1941).

Thorning, Graham, and Levine (1943a, 1943b) and Kerlin and Graham (1944a, 1944b) reported that fowl pox vaccines prepared from the entire chick embryo possess immunogenic properties.

Brandly and Dunlap (1939) present conclusive evidence that egg-propagated vaccines represent a distinct and desirable refinement over chicken-propagated vaccines. The pox-infected chorio-allantoic membranes were invariably richer in virus than the skin-lesion tissue of a similar age and handling; there was no apparent reduction of the pathogenicity or virulence of the virus for embryonated chicken eggs or chickens; pox viruses were easily propagated in embryonated chicken eggs; and egg-propagated vaccines were not contaminated with bacteria which may be found in chicken-propagated vaccines and may be of possible serious consequence for post-vaccination disturbances.

*Pigeon pox vaccine.* Pigeon pox vaccines are of two types: "pigeon propagated" and "chick-embryo origin" or "egg-propagated," depending upon the method of preparation.

Various methods have been used by investigators for preparing pigeon pox virus for vaccine. Graham and Brandly (1940) prepared vaccine by the application of a 1 per cent aqueous solution of powdered pigeon pox skin-lesion material to a defeathered area of the ventral surface of the breast in pigeons. The inoculated pigeons were killed when in a moribund condition, usually about the sixteenth day, at which time the affected skin and scab were removed. The material was desiccated, cut into small pieces, ground to a fine powder, distributed in suitable containers, and stored under refrigeration. When the virus is to be used, it is suspended in a 1 per cent solution in a suitable diluent such as sterile distilled water, physiological saline, or 20 per cent glycerin.

The "chick-embryo origin" or "egg-propagated" vaccine is prepared by propagation of the virus on the chorio-allantoic membrane of embryonated chicken eggs. The infected membranes are collected and processed as previously described for fowl pox vaccine. A suspension of at least 80 mgs. of virus in 4 cc. of diluent for the vaccination of 100 chickens is recommended by Beaudette (1941).

*Vaccination.* Fowl pox vaccine is used to vaccinate chickens and turkeys, and it may also be used for pheasants. Fowl pox vaccine is not to be used on pigeons, and it is never to be used on laying birds.

Pigeon pox vaccine is used to vaccinate chickens, turkeys, pigeons, and pheasants. It is used on chickens and turkeys when these birds are laying or when the flock is debilitated through the presence of other diseases or improper nutrition and management.

Fowl pox vaccines of adequate potency contain the causal agent of the disease and when improperly used are capable of producing the disease in a severe form in chickens. When properly used fowl pox vaccine is equivalent to a mild attack of the disease. Vaccination with fowl pox vaccine differs from the natural disease only in that the vaccine is applied to a small area, the extent of the lesion is less than that encountered in the natural disease, and vaccination is done at an age of the chicken when post-vaccination reactions are less likely to occur than in the natural disease.

All vaccine should be mixed away from the poultry house and precautions should be taken not to spill any of it on the premises. The vaccine should be mixed just before it is to be used and only enough prepared for one day's operation. If it is necessary to hold vaccine over night, it should be kept in the freezing compartment of an electric refrigerator. After the vaccine is mixed the hands should be washed to prevent contamination of the birds when they are handled since the vaccine should be kept away from all parts of the bird except the site of vaccination. When vaccination is completed the unused vaccine and all containers should be burned. The "stick" instruments should be boiled if they are to be kept for further use.

Since the flock will have to be examined for "takes," it is well to be consistent in the site of vaccination so that no confusion will occur in a false interpretation of the reaction to the vaccine.

Chickens may be vaccinated with fowl pox vaccine by one of two methods: the "stick" method and the "feather follicle" or brush method, since fowl pox virus has a predilection or tissue affinity for cutaneous epithelium and follicular cells. Chickens may be vaccinated with pigeon pox vaccine by the "feather follicle" method only, since pigeon pox virus has a particular affinity for follicular cells.

In the "stick" method the vaccine is introduced into the cutaneous epithelium by sticking the skin with a sharp-pointed instrument which has been moistened with the vaccine. The sites of application may be the skin of the leg, the under surface of the web of the wing, or on the breast. The undersurface of the web of the wing is probably the most convenient area for this type of vaccination. In baby chicks the vaccine is introduced into the skin of the flank region. The vaccinating instrument may be of any type which will insure adherence of the virus for its introduction into the skin. A sharp-pointed scalpel or knife with a narrow strip of adhesive tape wrapped about the blade so that only about $1/8$ inch of the point protrudes beyond the wrappings makes a satisfactory instrument. The object of the tape around

the blade is to prevent the blade from penetrating too deeply and causing an unnecessarily large wound.

Two medium-sized sewing machine needles or darning needles attached by the blunt ends to a small stick of soft wood from 4 to 6 inches long and about $\frac{3}{8}$ inch in diameter make a convenient instrument. The needles should be as nearly parallel as possible throughout two planes and slightly less than $\frac{1}{4}$ inch apart. The points should project out from the wooden handle about $\frac{1}{4}$ inch. The needles may be inserted into the handle by making two small holes with a fine wire brad and then forcing the blunt ends of the needles into the handle with a pair of pliers. A similar instrument may be made by inserting the blunt ends of the needles into a small cork.

Another satisfactory type of vaccinating instrument can be prepared by cutting on the transverse plane through the eye of a large sewing machine needle so as to leave two sharp points. The vaccine will adhere to the space between the points and the groove of the shank of the needle and will insure the deposit of a satisfactory amount of vaccine. The needle may be attached to a wooden handle or cork as described above.

When the "stick" method is used the vaccine should be in a container with a mouth wide enough to allow entrance of the instrument, so that the points can be moistened easily with the vaccine. A small ointment jar will be satisfactory unless the instrument used for this method can be made to fit the diluent bottle supplied with the vaccine. As an added precaution to prevent spilling of the vaccine, a small piece of wood, 2 inches by 4 inches by 6 inches long with holes large enough to receive the container of vaccine, should be prepared.

The stick instrument should be moistened with vaccine before each application.

With the "feather follicle" method the vaccine is applied to defeathered follicles with a brush. The brush should be stiff enough to withstand repeated usage without becoming moplike, and the bristles should be of a suitable length and number. Most manufacturers supply a brush with the vaccine. When fowl pox vaccine is applied by this method only three or four feather follicles should be infected as the reaction of the vaccine increases in proportion to the number of follicles infected. Usually, the most convenient vaccination area is the anterior or lateral aspect of one leg about midway between the hock joint and the femoro-tibial articulation of the leg. The bristles of the brush should be moistened with vaccine before each application and directed into the open follicles for proper deposition of the vaccine.

When pigeon pox vaccine is used to vaccinate chickens the vaccination area should be considerably larger than that for vaccination with fowl pox vaccine since the degree of immunity produced is in direct proportion to the size of the vaccination lesion, and pigeon pox virus does not produce a

systemic reaction in chickens. The feathers should be plucked from an area of about 1 x 2 inches on the leg and the exposed follicles over the entire area infected with the vaccine by directing the brush against the openings of the follicles. This is of particular importance in view of the affinity of pigeon pox virus for follicular cells.

The method of vaccination is a matter of personal preference. There are certain distinct advantages of the "stick" method over the "follicle" method with fowl pox vaccine which make the former method more desirable. According to Johnson (1934) some advantages of the "stick" method are as follows: uses less virus; permits more rapid vaccination; requires less help by eliminating feather plucking; results in less contamination of fowls and premises; results in less virus at "take"; standardizes vaccination procedure; results in less exposure of the "takes"; and provides a method applicable to fowls from a day old to maturity.

Reaction to fowl pox vaccine and evidence of a successful vaccination consists of a scab or "take" which may be readily detected in a week at the site of vaccination by either the "stick" or "feather follicle" method. The flock should be examined for "takes" between the sixth and tenth day following vaccination. If the vaccine was applied to the web of the wing by the "stick" method two small scabs will be observed on the under surface of the wing where the points of the vaccinating instrument were introduced into the skin and usually two small scabs on the outer surface of the wing where the points emerged (Fig. 25.5).

With the "feather follicle" method of vaccination there will be a swelling of each follicle and the formation of a scab (Fig. 25.6). These scabs will enlarge during the course of a week or 10 days and may coalesce to form a single scab covering the entire vaccination area.

A "take" following vaccination with pigeon pox vaccine is indicated by a swelling of the follicles which may be evident as early as the fifth day. Scabs are not produced in chickens immunized with this vaccine. A flock examination for "takes" should be made at about 10 to 12 days following vaccination. "Takes" with both fowl and pigeon pox vaccines subside and disappear about the third week following vaccination. The scabs dry and drop off the birds immunized with fowl pox vaccine.

The immunity developed in chickens following vaccination with fowl pox vaccines is enduring throughout the life of the chicken. With pigeon pox vaccine the duration of the immunity is not as long or as well established as that from fowl pox vaccine.

Vaccination provides no protection during the first two or three weeks, and during this period birds may become infected with the natural disease. Maximum immunity is attained by the end of the fourth week. Vaccination is only a prophylactic measure and should not be used for treatment of in-

fected chickens. When vaccination is to be done in a flock in which the disease has just made its appearance, all visibly affected birds should be removed from the flock and isolated to prevent spread of the infection from this source. Medicinal treatment of chickens against fowl pox is of no value.

When properly applied, a vaccine of adequate potency should produce "takes" in all of the vaccinated birds within the time previously specified for post-vaccination examination of the flock. Failure to obtain "takes" may be the result of the application of a vaccine of inadequate potency (use after



FIG. 25.5. Web of wing of chicken. Six-day "takes," fowl pox vaccine, stick method. (Brunett, Cornell Vet.)
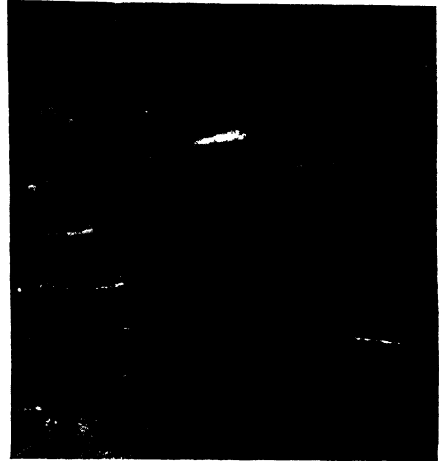
FIG. 25.6. Leg of chicken. Six-day "takes," fowl pox vaccine, brush method. (Brunett, Cornell Vet.)

expiration date, subjection to deleterious influences), improper application of the vaccine, or use on fowl pox immune birds. In the event that "takes" are not obtained in all vaccinated birds (except those previously immunized or recovered) the flock should be revaccinated at once.

The age of the bird at the time of vaccination with fowl pox vaccine is important. In certain areas day-old chicks are vaccinated, but the severe systemic reactions and high mortality rate following vaccination do not make birds of this age satisfactory subjects for vaccination. The preferable age for vaccination of chickens is from the sixth to the twelfth week according to Delaplane (1943) and from the eighth to the tenth week according to Beaudette (1941). As a rule the chicken should be at least one month old. Chickens should not be vaccinated within one month and preferably two months before production is expected to start. This allows ample time for the birds to recover from the effects of the vaccination before they reach the laying age. The upper limit for light breeds would be from three to three and one-half months and for heavy breeds from four to four and one-half months.

With pigeon pox vaccine the upper age limit for vaccination is not important since this vaccine does not produce a systemic reaction in chickens following its application. In young chickens, however, the age limit depends upon the feathering of the bird since the vaccine is applied only by the "feather follicle" method. Generally, chickens should be at least six weeks old, and it is preferable to have all of the birds in the flock well feathered before vaccination.

When pigeon pox vaccine is applied to pigeons only four or five follicles need to be infected, since this virus is as pathogenic for pigeons as fowl pox virus is for chickens. Follicles of either the breast or leg of the pigeon may be inoculated with the vaccine. The bristles of the vaccinating brush should be directed against the openings of the follicles to insure the deposition of the vaccine in the follicles. Pigeons should be vaccinated at about four to six weeks of age. Vaccinated pigeons should be segregated from non-vaccinated pigeons to prevent spread of the infection.

*Prophylactic vaccination.* Prophylactic immunization of chickens against fowl pox consists of vaccinating susceptible chickens with fowl pox vaccine prior to the time when the disease is likely to appear. Vaccination is usually done during the spring and summer months in those areas where the disease appears during the fall and winter months. In tropical climates where the disease may make its appearance throughout the year vaccination may be done at any time when warranted without regard to seasonal periods.

Vaccination is indicated in three types of flocks which present themselves in the problem of prophylaxis against fowl pox:

1. Vaccination is indicated as a routine prophylactic measure in a flock that has been infected with pox the previous year and the owner wishes to prevent such an occurrence in the new susceptible population. In this type of flock all young stock produced on the premises or introduced from other sources, since the outbreak, should be vaccinated with fowl pox vaccine. When several lots of birds are raised during the year, each lot should be vaccinated with fowl pox vaccine at the appropriate age as previously described. Vaccinated birds should be maintained under strict isolation as they are a source of infection for the nonvaccinated birds. If vaccination with fowl pox vaccine is delayed beyond a reasonable limit to expect the establishment of immunity prior to housing of the pullets, then pigeon pox vaccine should be used. This practice, however, should not be necessary.

2. This type of flock is one in which fowl pox was present the previous year but pigeon pox vaccine was used to check the spread of the disease at that time. Since pigeon pox vaccine does not induce a durable immunity in chickens the old birds should be revaccinated with fowl pox vaccine.

3. In certain congested poultry districts where fowl pox is prevalent, the flock owner should immunize the flock against pox through the application

of fowl pox vaccine to protect the flock against infection from the neighboring flocks. Delay under these circumstances may result in the infection being established at a time unfavorable for vaccination.

Prophylactic vaccination of pigeons with pigeon pox vaccine is indicated if the infection was present on the premises the previous year and if the loft is in a congested pigeon district.

Vaccination of canaries against pox with canary pox virus has not been successful, according to Burnet (1933a) and Durant and McDougle (1938). The latter authors observed that canaries recovered from the natural disease were refractory to further infection. Coulston and Manwell (1941) reported that canaries which had been vaccinated with the virus attenuated through storage did develop an immunity which protected them to some degree, but not completely, against exposure to fully virulent virus. As a therapeutic measure, 1 to 3 per cent mercurochrome in 70 per cent alcohol to which a trace of acetone was added was applied once or twice daily to the infected pox areas. The duration of the treatment varied with the severity of the disease. The authors concluded that "This method of treatment is successful in nearly all cases, unless they are very advanced. Recovered birds exhibit a strong immunity to reinfection, particularly if they have been infected to begin with by a virulent virus."

## REFERENCES

Antoniotti, D., and Romat, A.: 1910. Contribución al estudio del epithelioma contagioso del canario. Revista de Med. Vet. 22:326. Cited by Brunett (1915).

Barger, E. H., and Card, L. E.: 1943. Diseases and Parasites of Poultry. 3rd ed. Lea and Febiger, Philadelphia. P. 150.

Beach, J. R.: 1939. Report of committee on poultry diseases. Jour. Am. Vet. Med. Assn. 95:613.

Beaudette, F. R.: 1929. Some aspects of fowl-pox and its control. Jour. Am. Vet. Med. Assn. 75:563.

———: 1941. The reasons for failures in immunization against laryngotracheitis and pox. Proc. 45th Meet. U. S. Livestock Sanitary Assn., p. 127.

——— and Hudson, C. B.: 1938. Cultivation of pigeon-pox virus on the chorio-allantoic membrane. Jour. Am. Vet. Med. Assn. 93:146.

——— and Hudson, C. B.: 1941. Egg propagation of turkey pox virus. Poultry Sci. 20:79.

Bierbaum, K., and Gaede, H.: 1935. Die Züchtung von Geflügelpockenvirus in der Gewebekultur. Arch. wiss. u. prakt. Tierheilk. 69:441. Cited by Brandly and Dunlap (1938).

Blanc, G., and Caminopetros, J.: 1930. La transmission des varioles aviaires par les moustiques. Compt. Rend. Acad. Sci. France 190:954.

Bollinger, O.: 1873. Über Epithelioma contagiosum bein Haushuhn und die sogenannten Pocken des Geflügels. Arch. f. Path. Anat. u. Physiol. (Virchow) 58:349.

Borrel, A.: 1904. Sur les inclusions de l'épithélioma contagieux des oiseaux (molluscum contagiosum). Compt. rend. Soc. de biol. 2:642.

Brandly, C. A.: 1935. Some studies of infectious laryngotracheitis. Jour. Infect. Dis. 57:201.

———: 1936. Studies on the egg-propagated viruses of infectious laryngotracheitis and fowl-pox. Jour. Am. Vet. Med. Assn. 88:587.

———: 1937. Studies on certain filtrable viruses. I. Factors concerned with the egg propagation of fowl pox and infectious laryngotracheitis. Jour. Am. Vet. Med. Assn. 90:479.

———: 1941. Propagation of fowl- and pigeon-pox viruses in avian eggs and use of egg-cultivated viruses for immunization. Ill. Agr. Exper. Sta., Bul. 478.

——— and Bushnell, L. D.: 1932. Studies of some virus diseases of fowls. Jour. Am. Vet. Med. Assn. 80:782.

Brandly, C. A., and Dunlap, G. L.: 1938. An outbreak of pox in turkeys with notes on diagnosis and immunization. Poultry Sci. 17:511.

—— and Dunlap, G. L.: 1939. Studies on certain filtrable viruses. II. Immunization against fowl pox with fowl- and pigeon-pox viruses cultivated in vivo and in vitro. Jour. Am. Vet. Med. Assn. 95:340.

Brunett, E. L.: 1934. Some observations on pox virus obtained from a turkey. Rep. N. Y. St. Vet. Coll. (1932–33) :69.

——: 1945. Fowl pox. In Diseases of Poultry. 3rd printing. H. E. Biester and L. DeVries. The Iowa State College Press, Ames, Iowa. P. 481.

Burnet, E.: 1906. Contribution à l'étude de l'épithélioma contagieux des oiseaux. Ann. Inst. Pasteur 20:742.

Burnet, F. M.: 1933a. A virus disease of the canary of the fowl-pox group. Jour. Path. Bact. 37:107.

——:1933b. Unpublished. Cited by Burnet and Lush (1936).

——: 1936a. The use of the developing egg in virus research. Med. Res. Council. Special Rep. Series No. 220.

——: 1936b. Immunological studies with the virus of infectious laryngotracheitis of fowls using the developing egg technic. Jour. Exper. Med. 63:685.

—— and Lush, D.: 1936. The immunological relationship between Kikuth's canary virus and fowl-pox. Brit. Jour. Exper. Path. 17:302.

Carnwarth, T.: 1908. Zur Aetiologie der Hühnerdiphtherie und Geflügelpocken. Arb. a. d. kaiserl. Gesundheitsamt. 27:388. Cited by Doyle and Minett (1927).

Cary, C. A.: 1906. Chicken-pox, sore-head or contagious epithelioma in poultry. Ala. Agr. Exper. Sta., Bul. 136.

Coulston, F., and Manwell, R. D.: 1941. Successful chemotherapy of a virus disease of the canary. Am. Jour. Vet. Res. 2:101.

Dalling, T., Mason, J. H., and Gordon, W. S.: 1929. Fowl-pox antiserum. Brit. Jour. Exper. Path. 10:16.

de Blieck, L., and van Heelsbergen, T.: 1923. Impfung gegen Diphtherie und Geflügelpocken bei Hühnern. Deutsch. tierärztl. Wochenschr. 31:85.

Delaplane, J. P.: 1943. The differentiation of the respiratory diseases of chickens. R. I. Agr. Exper. Sta., Bul. 288.

Doyle, T. M.: 1930. Fowl pox. Rep. of Eleventh Internat. Vet. Cong. 3:675.

—— and Minett, F. C.: 1927. Fowl pox. Jour. Comp. Path. and Therap. 40:247.

Durant, A. J., and McDougle, H. C.: 1938. Investigation of pox in canaries. Proc. 42nd Meet. U. S. Livestock Sanitary Assn., p. 181.

Gallagher, B.: 1917. Epithelioma contagiosum of quail. Jour. Am. Vet. Med. Assn. 3:366.

Goodpasture, E. W.: 1928. Virus diseases of fowls as exemplified by contagious epithelioma (fowl-pox) of chickens and pigeons. In Filterable Viruses. T. M. Rivers. Williams and Wilkins Co., Baltimore. P. 235.

Graham, R., and Barger, E. H.: 1936. Studies on incubator hygiene. IV. A note on the virucidal effect of formaldehyde on fowl pox virus. Poultry Sci. 15:48.

—— and Brandly, C. A.: 1940. Immunization against pox in domestic fowl. Ill. Agr. Exper. Sta., Bul. 470.

——, Brandly, C. A., and Levine, N. D.: 1939. The effect of hard X-rays and ultra-violet light upon fowl pox virus in vitro. Cornell Vet. 29:383.

Green, R. H., Anderson, T. F., and Smadel, J. E.: 1942. Morphological structure of the virus of vaccinia. Jour. Exper. Med. 75:651.

Grosso. A. M., and Prieto. C.: 1939. Epitheliosis contagiosa de los canarios. Univ. de Buenos Aires. Instituto de Enfermedades Infecciosas. 1:No. 4. Cited by Brunett (1945).

Groupé, V., Oskay, J., and Rake, G.: 1946. Electron micrographs of the elementary bodies of fowl pox and canary pox. Proc. Soc. Exper. Biol. and Med. 63:477.

—— and Rake, G.: 1947. Studies on the morphology of the elementary bodies of fowl pox. Jour. Bact. 53:449.

Guarnieri, G.: 1892. Ricerche sulla patogenesi ed etiologia dell' infezione vaccinica e variolosa. Arch. sci. med. 16:403. Cited by Goodpasture (1928).

Hutyra, F., Marek, J., and Manninger, R.: 1938. Special Pathology and Therapeutics of the Diseases of Domestic Animals. Vol. I, 4th English ed. Alexander Eger, Chicago. P. 380.

Irons, V.: 1934. Cross-species transmission studies with different strains of bird pox. Am. Jour. Hyg. 20:329.

Johnson, E. P.: 1938. An unusual outbreak of chicken-pox. Jour. Am. Vet. Med. Assn. 93:115.

Johnson, W. T.: 1927. Fowl-pox prevention by immunization. Jour. Am. Vet. Med. Assn. 71:750.

———: 1934. Fowl pox. Rep. Twelfth Internat. Vet. Cong. 3:219.

Kerlin, D. L., and Graham, R.: 1944a. Studies on certain filtrable viruses. VI. Antigenic properties of entire embryo fowl pox vaccine. Proc. Soc. Exper. Biol. and Med. 55:225.

——— and Graham, R.: 1944b. Studies on certain filtrable viruses. VII. Antigenic properties of entire embryo fowl pox vaccine. Proc. Soc. Exper. Biol. and Med. 57:259.

Kikuth, W., and Gollub, H.: 1932. Versuche mit einem filtrierbaren Virus bei einer übertragbaren Kanarienvogelkrankheit. Zentralbl. Bakt. Abt. I. Orig. 125:313.

Kligler, I. J., and Ashner, M.: 1929. Transmission of fowl pox by mosquitoes: further observations. Brit. Jour. Exper. Path. 10:347.

———, Muckenfuss, R. S., and Rivers, T. M.: 1929. Transmission of fowl-pox by mosquitoes. Jour. Exper. Med. 49:649.

Ledingham, J. C. G.: 1931. The aetiological importance of the elementary bodies in vaccinia and fowl-pox. Lancet 221:525.

———: 1932. The development of agglutinins for elementary bodies in the course of experimental vaccinia and fowl-pox. Jour. Path. Bact. 35:140.

Loewenthal, W.: 1906. Untersuchungen über die sog. Taubenpocke (Epithelioma contagiosum). Deutsch. med. Wochenschr. 32:678.

McCulloch, E. C.: 1945. Disinfection and Sterilization. Lea and Febiger, Philadelphia.

McGaughey, C. A., and Burnet, F. M.: 1945. Avian pox in wild sparrows. Jour. Comp. Path. and Therap. 55:201.

Marx, E., and Sticker, A.: 1902. Untersuchungen über das Epithelioma contagiosum des Geflügels. Deutsch. med. Wochenschr. 28:893. Cited by Goodpasture (1928).

——— and Sticker, A.: 1903. Weitere Untersuchungen über Mitigation des Epithelioma contagiosum des Geflügels. Deutsch. med. Wochenschr. 29:79. Cited by Goodpasture (1928), Burnet (1906).

Matheson, R., Brunett, E. L., and Brody, A. L.: 1932. The transmission of fowl pox by mosquitoes, preliminary report. Rep. N. Y. St. Vet. Coll. (1930–31): 177.

Morosow, M. A.: 1926. Die Färbung der Paschenschen Körperchen durch Versilberung. Zentralbl. Bakt. Abt. I. Orig. 100:385.

Reis, J., and Nobrega, P.: 1937. Sobre um virus tripathogenico de bouba de canario. Arch. do Instituto Biologica 8:211. São Paulo, Brazil. Cited by Brunett (1945).

Rivolta: 1869. Cited by Goodpasture (1928) and von Reischauer (1906).

Robbins, B. H.: 1944. Effect of penicillin and patulin on fowl pox. Proc. Soc. Exper. Biol. and Med. 57:215.

Stafseth, H. J.: 1931. Pigeon-pox in Michigan. Jour. Am. Vet. Med. Assn. 79:822.

Stuppy, C.: 1932. Uebertragung von Geflügelpocken durch Mücken. Deutsch. tierärztl. Wochenschr. 40:260. Cited from Biol. Abst. (1934).

Syverton, J. T., and Cowan, I. M.: 1944. Bird pox in the sooty grouse, *Dendragapus fuliginosus fuliginosus* with recovery of the virus. Am. Jour. Vet. Res. 5:215.

te Hennepe, B. J. C.: 1926. Thèse pour le Doctorat Vétérinaire. Cited by Doyle and Minett (1927).

———: 1927. Combating poultry diseases by the State Serum Institute. Data from six thousand autopsies. Rep. Proc. Third World's Poultry Cong., p. 261.

Thorning, W. M., Graham, R., and Levine, N. D.: 1943a. Studies on certain filtrable viruses. IV. Immunogenic properties of fowl pox virus prepared from the entire embryo. Poultry Sci. 22:287.

———, Graham, R., and Levine, N. D.: 1943b. Studies on certain filtrable viruses. V. The immunogenic properties of the entire chick embryo inoculated with fowl-pox virus. Am. Jour. Vet. Res. 4:250.

von Reischauer, O.: 1906. Ueber die Pocken der Vögel ihre Beziehungen zu den echten Pocken und ihren Erreger. Zentralbl. Bakt. Abt. I. Orig. 40:356.

Ward, A. R., and Gallagher, B. A.: 1920. Diseases of Domesticated Birds. The Macmillan Co., New York. P. 96.

Woodruff, A. M., and Goodpasture, E. W.: 1931. The susceptibility of the chorio-allantoic membrane of chick embryo to infection with the fowl-pox virus. Am. Jour. Path. 7:209.

Woodruff, C. E., and Goodpasture, E. W.: 1929. The infectivity of isolated inclusion bodies of fowl-pox. Am. Jour. Path. 5:1.

——— and Goodpasture, E. W.: 1930. The relation of the virus of fowl-pox to the specific cellular inclusions of the disease. Am. Jour. Path. 6:713.

# CHAPTER TWENTY-SIX

## FOWL PEST

*By* E. L. STUBBS, *Department of Pathology, School of Veterinary Medicine,*
*University of Pennsylvania, Philadelphia, Pennsylvania*

☆     ☆     ☆

Fowl pest (*fowl plague, peste aviaire, Geflügelpest*) is an acute, highly infectious, generally fatal virus disease of fowls and sometimes of water birds.

**History.** Fowl pest was described first by Perroncito (1878) in Italy, and later studied by Rivolto and Delprato (1880), who found it to be different from fowl cholera and called it *typhus exudatious gallinarum.* Subsequently, it was discovered in enzootic form in southern Europe and has been observed in many countries of the world. It spread to Tyrol and southern Germany in 1898, and became common throughout Germany after the Brunswick poultry exhibition in 1901. It has been widespread in Austria, has been found in Switzerland, Rumania, and Russia, and occasionally has spread to France, Holland, and Great Britain. The disease is indigenous in Egypt, has extended widely in Asia, particularly in China and Japan, and has occurred also in South America.

Fowl pest was reported first in North America in 1924–25 and again in 1929. The first severe losses appear to have occurred in the poultry market of New York City, and others later in the poultry markets of New Jersey and Philadelphia. Dr. John R. Mohler, Chief of the United States Bureau of Animal Industry, reported the existence of the disease in the United States in December, 1924. Its occurrence in New York was reported by Brunett (1925) and in Pennsylvania by Stubbs in the same year. Later in 1925, Beaudette reported the presence of fowl pest in New Jersey; Julien, in Indiana; Boughton and Tunnicliff, in Illinois; and Johnson, in Michigan. An outbreak, reported by Beaudette and associates in New Jersey in 1929, was confined to a few flocks in one locality and was eradicated promptly. Hirt (1942) describes the appearance of fowl pest in Hungary after an absence of twenty-seven years. The disease spread from the west to the east throughout Hungary.

**Etiology.** Centanni and Savonuzzi (1900) demonstrated the cause of fowl pest to be a filtrable virus, and later their results were confirmed by other workers. Weineck (1940a) reports that the infectivity of tissues containing fowl pest virus is destroyed by extraction with alcohol ether, alcohol,

benzene, and chloroform but not by acetone or ether. Weineck considers this as evidence that the virulent component of the virus is lipoid in nature. The disease attacks chickens and related species, but chickens and turkeys are found affected most frequently and are considered most susceptible. With few exceptions, which seem to be immune, chickens are infected easily by subcutaneous, intramuscular, intraperitoneal, or intravenous injections of amounts even as small as one-millionth of a cubic centimeter. Feeding infection also succeeds. Introduction of the virus through injuries to the skin or by instillation into the eye produces the disease. After such infection, chickens die in 36 to 72 hours, and may or may not show symptoms.

Natural infection among pigeons is not found so commonly; water birds, such as ducks and geese, often remain free when chickens are attacked severely. It is a curious fact in this disease that it is confined frequently to a single species, and that other fowls on the same premises are not infected. Artificial infection, even by large amounts of virus from the species in which it has occurred naturally, often fails when injected into other species. Thus, on premises where there is great mortality among chickens, waterfowl usually are resistant. Injection of the virus into the central nervous system succeeds more frequently among pigeons and water birds, when the disease usually gives rise to nervous symptoms, such as convulsions and paralysis.

Mammals are considered immune, as artificial infection has been unsuccessful, and it is believed that this disease is not dangerous to man as there are no recorded cases. Morcos (1946) reports the successful transfer of the virus of fowl plague from birds into mammals by using defibrinated infected fowl blood injected subdurally into white mice in which the virus became fixed after the fifth passage. The intracerebral injection was made just above and posterior to the eye, and after the fifth passage produced death in about 3 days. At the same time, the virus became attenuated in fowls with a period of incubation of 6 days instead of the previous 3-day period. The mouse brain virus treated with ether was antigenic, and Morcos believes it is promising as an immunizing agent.

**Symptoms.** Fowl pest appears with sudden onset. Chickens may die without showing any symptoms. Usually there is weakness and an inclination to stay on the roost or in some secluded place to avoid disturbance. Dullness and inappetence are present. Hens stop laying, the feathers are ruffled, and the birds stagger (Fig. 26.1).

Cyanosis develops, with the comb and wattles becoming dark red or blackish. The eyes are dark, the eyelids close, the conjunctiva is red and swollen. There is fever, the body temperature rising to 110–112° F., and gradually lowering until it is subnormal, when it recedes to 100–103° F. Edema of the head, consisting of an exudation of serum into the subcutaneous tissues, marked around the eyes, ear lobes, and wattles, with a tendency to extend downward along the throat toward the breast, frequently appears.

Edema of the glottis may develop, followed by difficulty in respiration. The chickens may open their beaks for air and breathe with a rattling sound; suffocation may occur (Fig. 26.2). Mucus exudes from the nostrils, with a gray or reddish, blood-tinged exudate, which may cause the chickens to shake their heads in an effort to expel the obstructive discharge. Similar exudate may be found in the pharynx, which may contribute to the gasping and rattling sounds. The mucosa of the mouth may show small hemorrhages,



FIG. 26.1. Fowl pest. Dullness, listlessness, ruffled feathers.

and fibrinous exudate may be observed. Diarrhea also may be present, usually profuse and watery. Finally, the head cannot be raised from the ground, coma develops, the respiration becomes more labored, and death results, usually within 2 days.

Many modifications of symptoms may be noticed. Nervous disorders frequently are associated, particularly in cases that do not die early, with excitation, convulsions, rolling, or circling movements. There also may be ataxia and blindness. Jungherr, Tyzzer, Brandly, and Moses (1946) report that in experimental work with the Dutch East Indies strain of virus, the first symptoms were usually observed within 18 to 30 hours after inoculation with 100 or more minimal lethal doses of virus. Death occurred within the next 24 hours. The first evidence of the disease was a decrease of sensitivity to sensory stimuli. There was a pronounced general malaise, congestion of comb and wattles, and closing of the eyes, with the head resting on the breast or on the floor of the cage. Loss of appetite was evident, while the desire to drink remained so that birds frequently would fall asleep while drinking and let the water run from the mouth. Whether birds recovered or whether death occurred later, small focal or confluent areas of necrosis often were found on the comb and wattles. Inappetence, marked dehydration, loss of flesh, and in some cases severe torticollis with starvation were found. The corneas showed focal or diffuse opacities in rare cases.

**Pathology.** The virus of fowl pest is present throughout the body and in

the blood, the nervous system, all the tissues and tissue fluids, is secreted from the glands, and is found in the nasal and oral secretions, the intestinal and urinary excretions, and in the mucous and serous exudates. The virus, or materials containing it, is highly infectious in very small amounts; and the blood is so rich in virus that 0.000,000,1 cc. may be infectious for the chicken. The virus appears to be in close contact with the blood cells, while plasma or material free of cells is less infectious. It is killed easily; exposure to direct sunlight or to a temperature of 70° C. for a few minutes renders it inactive.



FIG. 26.2. Fowl pest. Chicken's head, swollen about eyes, wattles, and ear lobes.

Its activity is destroyed quickly by the common antiseptics and disinfectants. Cold, however, promotes longevity of the virus, and it may be preserved for long periods under refrigeration. Desiccation or glycerination preserves the virus so that it retains its virulence for years. Blood from chickens suffering from the disease, preserved in sealed test tubes, remains infectious for a long time. Filtrates retain their virulence with less uniformity than unfiltered material. Purchase (1931) showed that the virus of fowl pest retained its activity in flesh for 287 days, and in the bone marrow for 303 days when kept at chilling temperature. He believes the disease may be spread by feathers since he found that the virus survived on feathers for 18 days after being plucked from a chicken dead of fowl pest. Burnet and Ferry (1934) have reported the propagation of the virus by inoculation of the chorio-allantoic membranes of developing chick embryos. Jungherr et al. (1946) studied the inoculation of the Dutch East Indies strain into embryonating chicken eggs. Death was caused, and regardless of age or route of inoculation bright to dark red discoloration indicative of congestion of the embryonic tissues, especially the skin and less so of the musculature, occurred. Scattered punctiform hemorrhages were found in the skin and skeletal musculature with renal congestion in 12-day-old or older embryos. Minimum doses of virus in some eggs caused slower death of embryos which showed tumefaction and sometimes pinhead-sized, gray focal areas in the spleen and rarely in the liver. Variant viruses were obtained from such spleens which in further egg passages produced congestive and hemorrhagic lesions similar to the parent strain.

Histopathological studies were made on the embryonating chicken eggs. Inoculation into the allantoic sac usually failed to show any specific lesions. Inoculation of the virus onto the chorio-allantoic membrane was followed in

about one-fourth of the cases by shallow hemorrhagic ulcers in the ectoderm, filled with disintegrated red cells and heterophils, and delimited by an intensely congested and fibrotic zone of the mesoderm. The lesions in the embryo were multiple capillary hemorrhages, particularly in the skeletal muscles and in the spinal cord and brain as well as the myocardium and the gizzard wall.

Weineck (1940b) reports that pretreatment of 12-day-old chick embryos with methylene blue or neutral red 12 to 15 hours before infection, prevents death. Treatment with the dye 4 to 10 hours before infection only delays the infection which normally requires 12 to 14 hours to kill the embryo. Treatment with the dye 1 to 4 hours before injection had no effect, and so it was concluded that the first 4 hours after infection is a period of virus dissemination after which rapid production follows.

The changes found at post-mortem examination are those of septicemia. The lesions cannot be depended upon entirely for diagnosis, but are somewhat characteristic and usually quite uniform. Rigor mortis sets in early and is complete. There is cyanosis of the head; the face, comb and wattles are dark; and the conjunctiva is swollen, often petechiated. The nostrils and beak show accumulation of mucus, frequently stained or streaked with blood.



Fig. 26.3. Fowl pest. Chicken's head showing swollen wattles.

There may be edematous swellings of the head; and the fluid, clear and straw-colored, may be most marked in the face, about the eyes, in the ear lobes, the wattles, or in the subcutaneous tissue of the neck and breast (Fig. 26.3).

Removal of the skin shows engorgement of all blood vessels. The flesh is red. Hemorrhages are widespread and vary from the smallest possible petechiae to ecchymoses. Those that are small and widely scattered may be overlooked easily, but are conspicuous when grouped, as in the proventriculus or over the abdominal fat. They may be found in any tissue, frequently in the muscles of the breast. Very distinct petechial hemorrhages, quite characteristic and appearing to have been sprayed on with an atomizer, are found on the inner surface of the sternum when the breast is removed. Hemorrhages usually are found in the fat about the abdominal cavity, and petechiae frequently are sprinkled over the fat tissue forming the bottom of

this cavity. Petechial hemorrhages also are particularly noticeable in the abdominal fat over the proventriculus, gizzard, and mesentery, and in the thoracic fat over the heart.

The most characteristic lesion is the hemorrhagic alteration in the proventriculus, which can be seen after the whitish mucus is washed off the mucous membrane. Such hemorrhages, usually ecchymoses, may be observed between the conical elevations or secreting glands of this portion of the stomach, and are more noticeable when they occur as bright red blotches on the mucous membrane where it becomes smooth to enter the gizzard (Fig. 26.4).



FIG. 26.4. Fowl pest. Hemorrhages of proventriculus (chicken).

Petechiae or ecchymoses also are seen in the gizzard after the rough membrane or cuticle has been removed. The intestine frequently shows hemorrhagic changes, especially in the duodenum. These hemorrhages, petechiae or ecchymoses on the serous or mucous coat, are accompanied by catarrhal enteritis. The hemorrhagic enteritis usually present in fowl cholera is not found in fowl pest and may be helpful in differentiating these diseases.

The liver, spleen, and lungs do not show much change. Congestion may be found, and also hemorrhagic fluid in the peritoneum and pericardium. Exposure to the air results in clotting, and in some cases fibrinous exudate is already present. This is spoken of sometimes as the exudative form. The ovary, when functioning, shows highly engorged blood vessels, especially in the large follicles. The oviduct often shows gray exudation, and the wall is swollen.

Microscopically, the chief changes are congestion and hemorrhages. Perivascular round-cell infiltrations have been noted. Some have described necrosis in brain tissue and the presence of bodies resembling intracellular inclusions.

Beaudette and associates, reporting the outbreak of fowl plagues in New Jersey in 1929, record the occurrence of vesicles on the comb and wattles of chickens artificially infected. These vesicles were observed in cases that ran longer than the usual course, and varied in size from a pinhead to 4–5 mm. in diameter. The same investigators also report the occurrence of edema of the feet and tibiometatarsal joint; and spots of violet color on the

scales of the shanks and feet, some as mere spots and others as blotches 4–5 cm. in length.

Jungherr, Tyzzer, Brandly, and Moses (1946) in experimental work with the Dutch East Indies strain report a variety of post-mortem changes. The acute cases showed congestive, hemorrhagic, and transudative changes. Congestion was evident in the skin, the comb and wattles, musculature, oropharynx, larynx, trachea, and the abdominal viscera. Punctiform hemorrhages were most often found in the coronary fat beneath the epicardium, especially the left auricle and around the roots of the large vessels. Intestinal hemorrhages were discrete, scattered along the serosa in the walls of the intestine and in the mucosa of the proventriculus, and in the region of the Peyer's patches and the cecal tonsils. Transudative changes were less frequent and showed moderate to severe edema of the lungs with congestion or hemorrhage and consolidation. Pericardial fluid that jellied on exposure to the air was sometimes found, and in a few cases subcutaneous edema of the hocks, feet, breast, neck, and head was present. Usually the spleen appeared small and anemic, often almost white except for stellate areas of capillary injection near the mesenteric attachment.

Jungherr et al., also studied the histopathological changes in over 100 experimental cases of different ages while experimenting with the Dutch East Indies strain. They considered the basic lesion roundish, but not sharply delimited, foci of necrosis of various organs. The young foci were recognized by acidophilic staining. The cellular architecture was at first undisturbed, but in well developed foci the tissue cells were swollen and vesicle-like, with the cell membrane prominent, and the nucleus small and marginated leaving a large cytoplasmic space containing eosinophilic granules or globules. The necrotic foci did not show pyknosis or karyorrhexis, did not become confluent, but remained scattered. The foci were rarely numerous but were found in a variety of the organs such as the spleen, lung, thymus, liver, gall bladder, kidney, heart, pancreas, proventriculus, intestine, comb and wattles, iris, and occasionally in the gonads. The spleen showed the highest incidence of necrotic foci, and it was estimated that more than three-fourths of the cases showed spleen involvement with the other organs in a falling order of frequency.

The necrobiotic foci in various organs were often accompanied by hemorrhages, congestion, and edema. Hemorrhages were found in the submucosa of the secondary pulmonary bronchi, in the alveolar tissue, in the proventricular mucosa, in the thyroid, subepicardium, myocardium, and elsewhere. The lungs often showed edema and variable degrees of congestion with some proliferation, capillary congestion, and edema, and occasionally fibrinous thrombi were found in the region of necrotic foci.

**Diagnosis** of fowl pest may be returned when an acute, plaguelike infec-

tious disease resembling fowl cholera is encountered, accompanied by cyanosis and edema of the head, and hemorrhages in the proventriculus, gizzard, and abdominal fat. In questionable cases, negative bacteriological examinations, negative results from inoculations into mammals, and filtrates producing typical symptoms with characteristic lesions in chickens, remove the doubt. Differentiation from other acute infectious diseases, particularly fowl cholera, is difficult. Both diseases present similar symptoms and lesions: rapid onset, cyanosis, prostration, diarrhea, high mortality, and especially hemorrhagic alterations. The losses from fowl pest are regular and extensive, while from fowl cholera the mortality is likely to be irregular. In the recorded natural outbreaks of fowl pest in this country, where chickens are associated with waterfowl, such as ducks and geese, the latter are not affected; while in fowl cholera, ducks and geese are highly susceptible and show fowl cholera in a marked and highly fatal form. European literature indicates that young geese sometimes are affected. In numerous post-mortems of fowl cholera, areas of focal necrosis appearing as whitish-yellow points are observed scattered over the liver. Such changes have not been found in fowl pest. The hemorrhages of fowl cholera more often are confined to the heart and intestine, while in fowl pest the hemorrhagic lesions are more likely to be scattered throughout the body. Fowl cholera usually is suspected where there is prostration of individual fowls, sudden death in large numbers, and particularly marked hemorrhages found at autopsy. Bacteriological examination soon eliminates fowl cholera where fowl pest is present. In cholera, culture examination is positive; inoculation into rabbits, mice, and pigeons produces death, and filtrates do not cause the disease. Where there is recourse to laboratory procedure in fowl pest, culture examination and inoculations into rabbits, mice, and pigeons are negative, while filtrates will produce the disease in chickens.

Newcastle disease may be found in chicks, in growing chickens, and in mature fowl. The disease in chicks frequently begins with gasping, wheezing, or coughing, and the same changes are also found in older birds. It spreads very rapidly and may go through an entire group in one or two weeks. Later many different kinds of nervous symptoms and paralysis may be found. Shivering, incoordination, convulsions, and chronic spasms of the head or neck and body have been noted. Twitching of the head or tail may be seen, with birds walking in circles, forward or backward. Alternating periods of excitation and depression may be found. Birds may stand motionless with the head drawn back and eyes fixed. The head may be drawn toward the ground. The presence of respiratory difficulty followed by nervous symptoms points to Newcastle disease. Mortality may be high in young birds and slight or none in mature birds. Layers show an abrupt drop in egg production followed by irregular shells, discolored shells, and soft shells. Varying lengths of time are required for resumption of production.

**Differential diagnosis.** Doyle in England has described a disease similar to fowl pest from which it is difficult to differentiate. He discovered the malady near Newcastle and named it "Newcastle disease." It also has been described in other places as "pseudo-fowl pest." The symptoms are quite similar, and there is high mortality, but the pathologic changes are not nearly so marked. It can be transferred readily by blood, brain, organ emulsion, oral discharges, and feces, as well as by filtrates of these materials. The hemorrhages present in this disease are not nearly so marked as in fowl pest, and the period of incubation is longer (about one week or more). Experimental infection by contact often does not succeed in fowl pest, whereas in Newcastle disease contact infection appears easier.

Apoplectiform septicemia and sleeping sickness cause symptoms and lesions similar to fowl pest but can be differentiated by the demonstration of a streptococcus in the blood stream. These diseases produce depression, staggering, prostration, coma, and death. Post-mortem examinations disclose hemorrhages and hemorrhagic discolorations which are rather widespread. There may be lung congestion and hemorrhage, and usually a hemorrhagic pericarditis. Peritonitis is frequent and also catarrhal or hemorrhagic enteritis.

Phosphorous poisoning produces hemorrhagic lesions in the proventriculus and may be confused with fowl pest. Phosphorus, highly poisonous to chickens, causes depression, weakness, trembling, thirst, and sometimes diarrhea, and may result in sudden death. Post-mortem examinations may show hemorrhages in the proventriculus, usually with erosions, and extending more deeply into the tissue than in fowl pest. In phosphorous poisoning there is usually severe enteritis, particularly in the upper portion of the intestine. If such poisoning is suspected, attention should be directed to the detection of the phosphorous vapor that may be noticed as a transient cloud when the crop, proventriculus, and gizzard first are opened. The contents also have the distinctive odor of phosphorus, and if such material is taken into a dark room or mixed with dilute acid, the characteristic phosphorous luminosity is seen.

Botulism may be attended by sudden onset and cause high mortality. It may attack a flock with overnight suddenness, but the clinical picture is so striking that it should not be confused with any other disease. Usually no lesions are found in botulism.

Edema of the wattles, usually an infection of one or both wattles, with listlessness, inappetence, and marked depression, may be confused with fowl pest. Some cases show a slight swelling, while in others the wattles become enormous and occasionally rupture. The swelling first contains an edematous fluid which gradually thickens and becomes caseated. The mortality is not high unless the disease reaches the sinuses or spreads systemically. It is frequently due to the organism of fowl cholera.

**Prognosis.** The course of fowl pest is quite rapid in chickens, which often live only a few hours. After artificial injection the fowls usually die in 36 to 72 hours. Death frequently takes place after a short struggle, and the victim often is found dead on its back. Occasionally recovery occurs, and such survivors are solidly immune.

**Epidemiology.** The virus of fowl pest is present in many European countries, and develops where conditions are favorable, which, fortunately, is not with too great frequency. There seems to be continuous incidence in Italy, generally in severe form. Large numbers of chickens are exported by Italy to nearby countries; consequently, the disease has spread over Europe repeatedly and occasionally to other parts of the world. At various times certain regions have suffered severe losses, the disease having attained greatest prevalence throughout Europe in 1901 and 1925. Fowl pest may be suppressed rapidly and disappears after a time. It rarely reaches devastating proportions.

During the outbreak of fowl pest in the United States in 1924–25, the disease was most prevalent during the holiday trade in poultry at Thanksgiving and Christmas, when it caused considerable loss in the large poultry markets, particularly in New York and Philadelphia. Thence it spread to a few farms, in most instances through the addition to home flocks of chickens purchased at market. It is a not uncommon practice to purchase fowls in the live poultry market to be taken home and fattened. The diseased bird is without doubt the most dangerous factor in the spread of fowl pest, and one or more infected chickens introduced into a healthy group are capable of causing an outbreak. Usually the newly purchased chickens die first, although this is not always the case since carriers are known to exist, which may be responsible for the spread of the disease. Even though the fowls are confined closely and have no contact with others, the infection spreads to nearby groups. In other instances it will not appear in other groups on the same premises. Losses may begin within 1 or 2 days after contact.

The virus is present in the eye and nasal secretions and in the excretions of the nose and mouth, in the feces, and in the urine. The feed, drinking water, and soil of the pen become contaminated, and the mechanical spread of the virus on the shoes of attendants is probable. Similarly, livestock dealers provide excellent opportunity for the spread of the disease as they travel from place to place. Chickens may ingest the virus with contaminated food and other substances picked up from contaminated soil, and the virus also may enter through the respiratory tract. Since the virus exists in the blood, many believe that vectors play a part in the natural spread of the disease through the activity of blood-sucking insects. It is a commonly held opinion that the fact that susceptible chickens in close association with infected chickens frequently do not contract the disease but on injection readily succumb, lends support to the theory that vectors are instrumental in transmitting fowl pest.

It is also a curious fact that healthy chickens, placed in uncleaned, undisinfected cages in which others have died of fowl pest, frequently do not contract the disease. This is attributed to rapid destruction of the virus except under conditions favorable to its existence.

Wild and semiwild birds that associate with farmyard flocks also may spread the disease. Thus, pigeons, sparrows, and similar birds under some circumstances may disseminate the infection. Fowl pest also may be spread by streams, because in certain instances it has been noted that chickens on farms lying downstream from infected areas have contracted the disease, presumably from the water.

**Methods of control and eradication.** Outbreaks of fowl pest in America always have been controlled by eradication, and methods employed to limit and destroy the infection. In many instances the disease was self-limiting inasmuch as entire flocks succumbed, leaving no survivors. Since the infection spreads most easily and rapidly through the intermingling of fowl, exposure to infected premises, coops, crates, and other containers and carriers, the best control procedure is the destruction of all birds in the infected flock and the disinfection of housing and equipment.

Outbreaks should be reported immediately to livestock sanitary authorities. Poultrymen should be warned against the addition of new fowls to their flocks. If additions are necessary, the newly purchased fowl, regardless of source, should be isolated until they have been proven healthy. Sick fowls should be destroyed, carefully examined, and carcasses burned or properly buried. Frequent, diligent cleaning of premises, coops, crates, and carriers, followed by thorough disinfection is essential.

It is fortunate that the outbreaks of fowl pest in the United States were recognized and measures for control instituted promptly, for it is perhaps the most fatal of fowl diseases, capable of causing such destruction of the poultry population as to be of economic importance in diminishing the food supply. The dangerous character of the disease warranted the radical methods employed in each outbreak; complete eradication was effected within a few months. Quarantines were imposed, embargoes placed, and poultry shipping restricted; slaughter, sanitation and disinfection of poultry markets also aided in the control program. The cessation of traffic in live fowl after the holiday season probably contributed to a marked diminution of cases. According to Dr. Mohler, federal disease restrictions were applied to the poultry industry for the first time in the United States during the 1924–25 outbreak. Federal and state employees supervised the cleaning and disinfection of 2,718 plants, 8,140 cars, 352,525 coops, and 124,997 pieces of miscellaneous equipment. From available data, Dr. Mohler estimates that the direct losses from fowl pest in this outbreak were not less than one million dollars and probably considerably more. The successful eradication of fowl pest in the

United States, therefore, may be recorded as a noteworthy achievement of the veterinary profession through the cooperation of the American people.

## REFERENCES

Barger, E. H., and Card, L. E.: 1941. Diseases and Parasites of Poultry. Lea and Febiger, Philadelphia. 386 pp.

Beaudette, F. R.: 1925. Observations upon fowl plague in New Jersey. Jour. Am. Vet. Med. Assn. 67:186.

———, Hudson, C. B., and Saxe, A. H.: 1934. An outbreak of fowl plague in New Jersey in 1929. Jour. Agr. Res. 49:83.

Boughton, I. B., and Tunnicliff, E. A.: 1925. European fowl pest in Illinois. Jour. Am. Vet. Med. Assn. 67:183.

Brunett, E. L.: 1925. The occurrence of a disease of chickens in New York State caused by a filtrable virus. Jour. Am. Vet. Med. Assn. 66:497.

——— and Kondo, S.: 1926. Fowl plague. Rep. N. Y. St. Vet. Coll. (1924-25). P. 209.

Burnet, F. M., and Ferry, J. D.: 1931. The differentiation of the viruses of fowl plague and Newcastle disease: Experiments using the technic of chorio-allantoic membrane inoculation of the developing egg. Brit. Jour. Exper. Path. 15:56.

Centanni and Savonuzzi: 1900. Cited by Gerlach, 1929, Kolle and Wass. Path. Mikr. 9:165.

Doyle, T. M.: 1927. A hitherto unrecorded disease of fowls due to a filter-passing virus. Jour. Comp. Path. and Therap. 40:144.

Freese, Dr.: 1908. Fowl-plague, with special reference to its pathological anatomy. Trans. from the Deutsch. tierärztl. Wochenschr., 1908, 173. Jour. Comp. Path. and Therap. 21:212.

———: 1925. Fowl pest with special consideration of the pathology of the disease. Trans. by L. P. Doyle. Jour. Am. Vet. Med. Assn. 67:203.

Gerlach, F.: 1929. Kolle and Wass., Path. Mikr. 165.

Hirt, G.: 1942. Pathological findings in fowl plague (trans. title). Abst. Deutsch. tierärztl. Wochenschr. 50:453. (Abst. Vet. Bul. 13:388.)

Hutyra, F., Marek, J., and Manninger, R.: 1938. Special Pathology and Therapeutics of the Diseases of Domestic Animals. Alexander Eger, Chicago, Vol. 1.

Johnson, S. R.: 1925. European fowl pest in Michigan. Jour. Am. Vet. Med. Assn. 67:195.

Julien, R. C.: 1925. Fowl pest in Indiana. Jour. Am. Vet. Med. Assn. 67:178.

Jungherr, E. L., Tyzzer, E. E., Brandly, C. A., and Moses, H. E.: 1946. The comparative pathology of fowl plague and Newcastle disease. Am. Jour. Vet. Res. 7:250.

Krohn, L. D.: 1925. A study on the recent outbreak of fowl disease in New York City. Jour. Am. Vet. Med. Assn. 67:146.

Lerner, — and Wojtek, —: 1942. Hühnerspirochätose. Deutsch. tierärztl. Wochenschr. 50:364. (Abst. Vet. Bul. 13:51.)

Matzke, M.: 1942. Die Diagnose der Hühnerpest. Zeitschr. Infekt-Krankh. parasitäre Krankh. u. Hyg. der Haustiere 59:42. (Abst. Biol. Abst. (1943) 17, No. 20580, p. 1948.)

Mohler, J. R.: 1924. Statement from Bureau of Animal Industry.

———: 1926. Fowl pest in the United States. Jour. Am. Vet. Med. Assn. 68:549.

Morcos, Z.: 1946. Fowl plague in Egypt. Immunization mouse neurotropic fixed virus. Vet. Jour. 102:3.

Perroncito: 1878. Cited by Gerlach, 1929, Kolle and Wass. Path. Mikr., p. 165.

Purchase, H. S.: 1931. Experiments on the viability of the virus of fowl-plague under trade conditions. Vet. Record 11:644.

Reis, J., Nobrega, P., and Reis, A. S.: 1936. Tratado de Doencas das Aves. Instituto Biologica, São Paulo, Brazil. 468 pp.

Rivolto and Delprato: 1880. Cited by Gerlach, 1929, Kolle and Wass. Path. Mikr., p. 165.

Stubbs, E. L.: 1925. Fowl plague. Univ. of Pa. Quart., 20.

———: 1925. Fowl plague in Pennsylvania. Jour. Am. Vet. Med. Assn. 67:180.

———: 1926. Fowl pest. Jour. Am. Vet. Med. Assn. 68:560.

———: 1946. Newcastle disease in Pennsylvania. Univ. Pa., Bul. 46:3.

van Heelsbergen, T.: 1929. Handbuch der Geflügelkrankheiten und der Geflügelzucht. Ferdinand Enke, Stuttgart.

Weineck, E.: 1940a. Ueber die Protein-Lipoid-Simplexnatur des Hühnerpestvirus. Zeitschr. f. Immunitätsforsch. 98:463. (Abst. Vet. Bul. 12:278.)

———: 1940b. Ueber die Reaktionsfähigkeit des Hühnerpestvirus nach quantitativ abgestufter Absättigung der Erythrozyten. Zeitschr. f. Immunitätsforsch. 98:469. (Abst. Vet. Bul. 12:278.)

# FOOT-AND-MOUTH DISEASE IN FOWL

By PETER K. OLITSKY, *Laboratories of The Rockefeller Institute for Medical Research, New York*

☆　☆　☆

**Susceptibility to natural disease.** The older textbooks (Hutyra and Marek, 1905; van Heelsbergen, 1929; Ehrhardt, 1914) refer to the observations of Spinola, Wildner, and of Becker on the clinical appearance of foot-and-mouth disease in fowl. Briefly, the descriptions include local and general reactions: A vesicular exanthem of small, scattered lesions is noted on the comb, the conjunctiva, about the nostrils, the wattles, and toes, along with a similar enanthem in the mouth and throat. After a few days the vesicles burst, leaving eroded areas which heal over within a week or two. The general reactions are fever and weakness, the latter being due to interference with feeding by the oral and throat erosions. Complete recovery ensues within one or two weeks—the disease is not fatal. No experimental work on identification of the causal agent is reported.

It is plain that clinical diagnosis, by itself, in the absence of laboratory investigations on the disease, is insufficient to label the described affection as foot-and-mouth disease. This is especially true in view of the known resistance of fowl to the usual strains of the cattle or guinea pig virus. Van Heelsbergen (1929) states that the described lesions resemble those of fowl pox, that he has often observed vesicular "eczema" on the comb and wattles of chickens that simulated the exanthem of fowl pox, and finally, that such lesions are seen in fowl deriving from areas free from foot-and-mouth disease. Reis and Nobrega (1936) state that the relationship between these manifestations as reported and foot-and-mouth disease is yet to be shown.

**Susceptibility to experimental inoculation with the virus.** Earlier attempts to induce an infection in fowl by inoculation or feeding virus have been unsuccessful (van Heelsbergen, 1929). Minett (1927), of the British Foot-and-Mouth Disease Committee, found that in three of twelve fowls fed large amounts of guinea pig virus, the latter was detected, by the calf inoculation test, in the feces passed between 10 and 24 and possibly 26 hours, but not later, after feeding. He also reported inability to induce apparent disease in "fowl" generally and in ducks, sea gulls, martins, and sparrows specifically, although later he was successful in producing local lesions in certain birds, as will be

described. Minett (1927) found, however, that in two of thirty-four fowl the virus could persist at the point of inoculation at the base of the claws for 5 days and in the breast muscle 2 days. With respect to ducks, the virus persisted for 3 days without producing lesions at the inoculation zone in the webs; but when Galloway (1937), of the same Committee, inoculated sixteen young adult ducks intradermally into the pads at the base of the feet and on the digits, thirteen showed vesicles 2 to 3 days later on the upper surfaces of the web. The vesicles harbored virus, and the disease was transmitted through eight consecutive duck passages. The virus was also detected in the blood in certain instances on the third day after injection, but no contact spread of the disease from duck to duck arose. In one of ten sea gulls, a single large vesicle was produced in the web by the same method, and the vesicular fluid contained virus.

The British investigators, who have conducted the most extensive researches on the susceptibility of birds to infection, conclude that there is some degree of susceptibility, but the results of their work generally give "no very striking support to the hypothesis of the spread of the disease by birds." However, with regard to sea gulls, which feed on farm land and travel great distances, the British observers state that the possibility of their becoming infected naturally is not wholly excluded, and the question of their occasional transportation of virus, mechanically, should also be considered.

Repeated field observations and experiences in the United States on past occurrences of foot-and-mouth disease have never indicated that birds have transmitted the infection from one premise to another (personal communication, S. O. Fladness, Chief, Field Inspection Division, Bureau of Animal Industry).

As a point of interest, it may be stated that several authorities (Kling et al., 1926, 1939; Waldmann and Hirschfelder, 1938; and others) regard man, of all living beings, the main agent in spreading the virus.

**Propagation of the virus in eggs.** Attempts by Galloway (1937) to propagate the virus of foot-and-mouth disease in the chorio-allantoic membrane of developing hen and duck eggs were not successful. Although the virus was found to survive in certain cases in the chorio-allantoic membrane of duck eggs, only two or three successful passages of virus from duck egg to duck egg were achieved, and no further passages were possible. Likewise, no evidence was obtained to prove that multiplication of virus had taken place. The foot-and-mouth disease virus can, however, be cultivated in tissue cultures in the presence of embryonic tissues deriving from susceptible species of animals [e.g., skin of embryo guinea pigs (Hecke, 1932)]. Recently its propagation has been achieved also in adult tissues.

**Diagnosis of the virus.** The diagnosis of a virus recovered from fowl, such as that of foot-and-mouth disease, is made by the cutaneous injection of

vesicular fluid in the scarified foot pads of healthy, adult guinea pigs. Twelve hours to 5 days later characteristic vesicles appear which are transmissible in series to normal guinea pigs. There are at present three immunologically distinct strains; according to German (Waldmann) classification they are A, B, and C; according to the French (Vallée) terminology, the A is called O and the B is called A, the C being a new strain. The strain of virus is determined by cross-immunity tests in guinea pigs, as well as by serological tests of complement-fixation and neutralization, with known types of antiserum or with suspected serum tested against standard strains of virus. Type specific antisera are ordinarily derived from convalescent or immunized guinea pigs.[1]

## REFERENCES

Ehrhardt, H. W.: 1914. Die Krankheiten des Hausgefiügels. Aarau E. Wirz, 3rd ed . Quoted by Ward, A. R., and Gallagher, B. A. 1926. Diseases of Domesticated Birds. The Macmillan Co., New York. P. 142.

Galloway, I. A.: 1937. Fifth Progress Report of the Foot-and-Mouth Disease Research Committee, H. M. Stationery Office, London. Pp. 29, 364, 369.

Hecke, F.: 1932. Die Eignung verschiedener Gewebsarten zur Zuchtung des Maul- und Klauen- seuchevirus. Zentralbl. f. Bakt. I. Orig. 125:321.

Hutyra, F., and Marek, J.: 1905. Spezielle Pathologie und Therapie der Haustiere. Gustav Fischer, Jena. I:307–8.

Kling, C., and Höjer, A.: 1926. Recherches sur le mode de propagation de la fièvre aphteuse. Transmission du contage. Compt. rend. Soc. de biol. 94:615.

————, Huss, R., and Olin, G.: 1939. Présence du virus de la fièvre aphteuse dans le contenu intestinal d'un sujet humain vivant dans un milieu infecté. Compt. rend. Soc. de biol. 131:478.

Minett, F. C.: 1927. Second Progress Report of the Foot-and-Mouth Disease Research Committee, H. M. Stationery Office, London. Pp. 18, 31, 50.

Reis, J., and Nobrega, P.: 1936. Tratado de Doencas das Aves. Instituto Biologico, São Paulo. P. 45.

van Heelsbergen, T.: 1929. Handbuch der Geflügelkrankheiten und der Geflugelzucht. Ferdinand Enke, Stuttgart. Pp. 308–10.

Waldmann, O., and Hirschfelder, H.: 1938. Die epizootische Bedeutung der Ratten, des Wildes, der Vögel und der Insekten für die Verbreitung der Maul- und Klauenseuche. Berliner tierärztl. Wochenschr. 54:229.

[1] It should be stressed at this point that the United States Department of Agriculture has, on good grounds, never permitted experimental work with the virus of foot-and-mouth disease in this country, nor the importation into it of the virus from abroad (see Jour. Am. Med. Assn., 1925, 84:1143) .

# RABIES AND INFECTIOUS EQUINE ANEMIA

*By* L. H. SCHWARTE, *Veterinary Research Institute,*
*Iowa State College, Ames, Iowa*

☆   ☆   ☆

## RABIES IN FOWL

Rabies is an acute infectious disease caused by a filtrable virus. It is characterized by symptoms of a central nervous disturbance followed by paralysis, terminating fatally in most cases.

Gibier (1884) was one of the first to conduct experimental investigations on rabies infection in fowl. He was successful in transmitting the disease to chickens and was able to reinfect mammals with virus recovered from diseased birds. He also reported cases of spontaneous recovery in experimentally infected birds.

Kraus and Clairmont (1900) carried on extensive investigations relative to the susceptibility of various species of birds to rabies as well as the variations in the course of the disease and in the clinical symptoms manifested. They reported that the raven, falcon, and old pigeons were refractory to rabies infection. However, the latter could be infected following a period of starvation. Young pigeons were found to be susceptible to rabies. A great variation in the incubation period was recorded from two weeks in owls and geese to 40 days or more in chickens. They also observed gradual cessation of symptoms and slow recovery in some instances. Vaccines made from avian blood sera or brain tissues were ineffective as immunizing agents. They were successful in transmitting rabies from birds to rabbits, but the incubation period was progressively extended until the virus was rendered inactive.

The clinical symptoms in the fowl included incoordination, paresis followed by paralysis, emaciation, and death. The characteristic lesions in the brain and spinal cord were similar to those found in man and animals. Chronic cases showed better developed lesions.

V. Löte (1904) reported that some of the birds of prey were susceptible to rabies infection. He infected a mouse hawk (*Buteo vulgaris*) subdurally with virus secured from the medulla oblongata of a rabbit. The bird refused to eat 11 days following inoculation and manifested symptoms of a central nervous disturbance. Later, convulsions of short duration were observed especially when the bird was aroused. On the third day following the first

appearance of clinical symptoms, it could no longer stand, and lay on its right side prostrated. The next day the bird was dead.

V. Löte also successfully transmitted rabies to two eagle-owls which died two and a half and nine months, respectively, subsequent to inoculation, without manifesting any appreciable clinical symptoms. He was able to transmit rabies to guinea pigs with brain tissue from these birds. V. Löte considered chickens and pigeons to be more resistant to artificial infection than birds of prey. He infected three cocks experimentally, only one of which contracted the disease. The course of the disease in this bird was rather unusual. After an incubation period of 43 days the first symptoms were manifested. The bird refused food and showed evidence of incoordination. Decided improvement was observed 3 days after the first appearance of clinical symptoms. For 14 days the subject appeared normal in every respect followed by severe paralytic symptoms. This condition lasted about one week followed by recovery. A similar course of this disease was observed in a hen which completely recovered.

Marie (1904) observed the great variation in the incubation period and course of rabies in birds. He attempted to increase the neutralizing power of the sera of mature pigeons which were apparently immune to rabies by the administration of large doses of active virus. These attempts were not successful. Repeated passage of "street" virus through birds resulted in decreased virulence and finally the inactivation of the virus to a point where it was no longer capable of producing any reaction in mammals. After serial passage of active virus through seven or more chickens, the administration of suitable quantities of brain emulsion intraperitoneally or subcutaneously protected mammals against intraocular inoculation of "street" virus.

Remlinger and Bailly (1936) experienced no difficulty in transmitting rabies to the stork (*Ciconia ciconia*) by intracerebral inoculation of "street" virus. The symptoms observed were exclusively of the paralytic type. Jacotot (1938) reported the experimental transmission of rabies to the pheasant (*Diardigallus diardi* B.P.).

Remlinger and Bailly (1929a, b) considered the occurrence of rabies in the chicken under natural conditions quite exceptional. They found, however, that the disease could be transmitted to the bird by bites inflicted on the comb by a rabid dog. Symptoms of the furious type or the paralytic form may develop after a short or comparatively long incubation period. In the furious form the diseased bird may attack its mates or other animals with its beak. They considered rabid chickens a potential source of danger in transmitting the disease to animals and man.

The literature contains very little definite proof of the occurrence of spontaneous rabies in fowls. Most of our information concerning the avian form of this disease has been secured by experimental investigation. Schweinburg

(1928), however, reported a case in which a patient had been injured by a rabid hen. The hen showed symptoms of the furious form of rabies for a period of 3 days.

## REFERENCES

Gibier, P.: 1884. Recherches expérimentales sur la rage. Abst. Compt. rend. Acad. d. Sc. 98:531.

Jacotot, H.: 1938. Transmission de la rage au faisan (*Diardigallus diardi* B.P.). Compt. rend. Soc. de biol. 127:131.

Kraus, R., and Clairmont, P.: 1900. Über experimentelle Lyssa bei Vögeln. Zeitschr. f. Hyg. 34:1.

Marie, M. A.: 1904. Note sur la rage chez les oiseaux. Compt. rend. Soc. de biol. 56:573.

Remlinger, P., and Bailly, J.: 1929a. La rage du coq. Ann. Inst. Past. 43:153.

———— and Bailly, J.: 1929b. Nouvelles observations relatives à la rage du coq. Bul. de l'Académie Vét. 82:286.

———— and Bailly, J.: 1936. Transmission de la rage à la cigogne (*Ciconia ciconia*). Compt. rend. Soc. de biol. 123:383.

Schweinburg, F.: 1928. Seuchenbekämpfung. Jahresber. vet. Med. 48:930.

v. Löte, J.: 1904. Beiträge zur Kenntnis der experimentellen Lyssa der Vögel. Zentralbl. f. Bakt. I. Orig. 35:741.

## INFECTIOUS EQUINE ANEMIA IN FOWL

Infectious equine anemia or swamp fever is a specific infectious disease caused by a filtrable virus. The transmissibility of this disease to birds has been a controversial question for a number of years. The evidence in support of its transmissibility to birds is not adequate to justify definite conclusions. However, the reports found in the literature on this subject are of interest and deserve consideration.

Oppermann and Lauterbach (1928) reported that chickens may be infected with the virus of infectious anemia of horses based on their observations and investigations. They observed histopathologic changes in the liver of birds similar to those found in infected horses. The infiltrations of round cells and the deposition of hemosiderin in the liver were believed to be significant. This disease could also be diagnosed in horses on farms where the chickens showed what Oppermann and Lauterbach considered typical liver changes of infectious anemia. They reported that it was possible to infect chickens experimentally with manure from horses that suffered from this disease and produce typical liver changes in them. If chickens were infected with the virus of infectious anemia, no definite clinical symptoms could be detected, but they considered the disease to assume a very mild form. These birds reacted to this infection by showing a reduction in the number of red corpuscles. They considered it probable that the hemosiderin deposition in the liver was the result of the destruction of the erythrocytes. Certain cases were also reported in which chickens died of the infection. The liver changes in the chicken were considered by Oppermann and Lauterbach as one of the most important criteria in the diagnosis of infectious equine anemia. These changes were found to be most consistent when the chickens were killed 5 to 7 days after infection. They concluded that since man is also

susceptible to infection by this virus, spontaneous infections of chickens are important in the control of this disease.

Balozet (1937a, b) was not able to confirm the work of Oppermann and Lauterbach. He was not able to transmit infectious equine anemia to chickens nor was he able to recover the virus from inoculated birds. He considered them completely refractory to the disease.

Gochenour, Stein, and Osteen (1938) after many years of investigation on the nature, course, and transmission of infectious equine anemia considered that this disease was confined largely to horses, mules, and asses. A few cases have been reported in man.

Stein (1940) inoculated the chorio-allantoic sac of 5- to 12-day-old chick embryos with virulent blood from subacute, chronic, and carrier cases of infectious anemia. No alterations in the appearance, condition, or development of the embryos occurred. Eleven-day-old chick embryos were also inoculated intravenously with virulent blood drawn from horses during a febrile attack. There was no evidence of untoward effects on the development of any of the embryos.

In view of the rather extensive investigations which have been carried on in recent years on infectious equine anemia, no conclusive evidence has been presented to indicate that this disease may be transmitted to birds. Apparently too much dependence has been placed on the histologic changes observed in inoculated birds. The absence of characteristic clinical symptoms and the failure to transmit the disease to the horse, mule, or ass by the actual inoculation of material from inoculated birds, leaves reasonable doubt as to the susceptibility of the avian species.

## REFERENCES

Balozet. L.: 1937a. Inoculation du virus de l'anémia infectieuse des équidés à d'autres espèces. Compt. rend. Soc. de biol. 124:1150.

———: 1937b. Etudes expérimentales sur l'anémic infectieuse des équidés. Arch. Inst. Past. de Tunis 26:27.

Gochenour, W. S., Stein, C. D., and Osteen, O. L.: 1938. Infectious anemia. U.S.D.A. Farmers' Bul. No. 1819.

Oppermann and Lauterbach: 1928. Die Diagnose der infektiösen Anämie des Pferdes mit Hilfe des Hühnerversuches. Deutsch. tierärztl. Wochenschr. 36: (Festschrift after p. 878) 61.

Stein, C. D.: 1940. Report of the Chief of the Bureau of Animal Industry.

# CHAPTER TWENTY-NINE

# AVIAN MONOCYTOSIS[1]

## (So-called Pullet Disease)

*By* ERWIN JUNGHERR, *Department of Animal Diseases,*
*University of Connecticut, Storrs, Connecticut*

☆ ☆ ☆

**Synonyms.** Pullet disease, blue comb, summer disease, housing disease, unknown disease, new disease, X disease (Beaudette, 1929), XX disease, cholera-like disease (Ryff and Stafseth, 1942), contagious indigestion (Waller *et al.,* 1942), battery nephritis, Bright's disease (Weaver, 1941), Tom Barron's disease, acute toxemia or colibacillosis (Weisner, 1941), hepato-nephrosis (Jungherr and Levine, 1941), avian monocytosis. (Jungherr and Matterson, 1944).

Under the term X disease Beaudette ·(1929) briefly described a disorder of adult fowl which usually affected heavy birds in high production and was characterized by cyanosis of the comb and wattles and sudden death; flock mortality was comparatively low. At autopsy affected birds showed congestion of the respiratory tract, liver, ovary, kidneys, and intestine, the intestine being filled with thick catarrhal material. The heart and the abdominal fat surrounding the gizzard showed small hemorrhages; in one case the liver exhibited evidence of necrosis. The disorder revealed an anatomic resemblance to fowl cholera, but culture and transmission studies with unfiltered and filtered materials failed to demonstrate an infectious agent.

During the past decade a similar condition of chickens and occasionally of turkeys has been observed both in the field and in the laboratory, throughout the northeastern states, but no systematic study has been reported until Jungherr and Levine (1940) attempted a pathologic delineation of the syndrome. Although these authors recognized an acute form similar to X disease, and a subacute form primarily characterized by kidney lesions, they found certain microscopic and chemical features to be common to both forms and regarded them as an entity. On purely symptomatic and gross-pathologic grounds, Bullis (1940) believed the acute and subacute forms, termed by him "pullet" and "blue comb" disease, respectively, to represent different entities. This possibility was likewise considered by Beaudette

(1940), who differentiated them and applied the names "X disease" and "new wheat poisoning."

These opinions emphasize the common occurrence in young laying birds of an important disorder which has the earmarks of an infectious disease but for which a transmissible etiologic agent has not been demonstrated with certainty. Without at least tentative recognition of "pullet disease" as a definite condition, certain cases of adult morbidity and mortality could not be diagnosed.

**Occurrence.** The statistical data on the geographic distribution of pullet disease are limited, because the disease has been recognized pathologically only in recent years. Lack of agreement on the morphologic range of the syndrome retards diagnostic classification. Some reports of the disease have been based on symptomatic evidence alone. However, even if one considers only the acute and most easily recognizable form, the occurrence of pullet disease has been reliably reported, aside from New Jersey (Beaudette, 1929), from most of the northeastern states, Michigan (Weisner, 1941), and more recently from California (Hurt, 1941) and Ontario (Weaver, 1941). Verbal reports seem to indicate its presence in North Carolina, Utah, and Missouri (reported by Jungherr, 1945). Gordon and Blaxland (1945) reported the occurrence in England of a disease in poultry resembling the so-called pullet disease in America.

What has been said in respect to the significance of geographic data holds true for other anamnestic observations. A Connecticut survey for the years 1931–39 (Jungherr and Levine, 1941) showed that pullet-disease-like conditions occurred in 15 per cent of 1,765 survey cases examined; 72 per cent of the positive cases were classified as uncomplicated. These cases were observed in flocks of birds kept on rations prepared according to a standard New England formula, and on twenty different commercial brands. This was of particular interest, since the diseased condition is of apparently metabolic origin. Various poultry breeds were found to be susceptible, the heavy breeds predominating. The majority of the cases occurred between the ages of five and seven months, that is during early production, but pathologically indistinguishable cases were observed in chicks four weeks old and in two-year-old layers. Turkeys occasionally showed the syndrome. The available data placed the major seasonal incidence between June and November, with the peak in August. A continuation of the Connecticut survey for the four calendar years 1940 to 1943 showed a similar seasonal distribution of the incidence (Jungherr and Matterson, 1944). During 1943 the attack rate was particularly high; namely, 34.5 per cent of 269 specimen consignments of chickens three months of age or older (Scott, Jungherr, and Matterson, 1944). Since that time the numerical incidence has decreased markedly, a fact for which no ready explanation has become available.

**Symptoms.** In the typical acute form a large proportion (average 15 to 21 per cent) of an apparently healthy flock shows a sudden affliction which is characterized by depression, lack of appetite, and whitish or watery diarrhea; occasionally there is constipation. Some birds exhibit distension of the crop with sour-smelling contents, darkening of the head (blue comb or cyanosis), sunken eyes, shrivelled legs, and high fever in the terminal stages. Laying flocks undergo a severe drop in egg production. Mortality is usually sudden and ranges from 50 per cent to almost zero, with an average of about 5 per cent of the flock. Subacute cases are distinguished by a comparatively low, often spotty, incidence and prolonged course. The clinical signs in the flock are less intense; but individual birds, according to Weaver (1941), may show severe prostration, oliguria, convulsive symptoms, and impaired vision. The primary symptoms of pullet disease are nonspecific in themselves, but when considered together with the seasonal incidence during early active production, they are highly suggestive of the disorder, if known infectious diseases can be ruled out. Sporadic cases often go unnoticed and are classed among the culls.

The course of the disease in most cases extends over a period of from one to two weeks, and terminates in a high percentage of apparent recovery, especially if prompt attention is given to the ailing flock. Egg production, however, tends to lag for several weeks, and a partial moult may ensue. After the acute attack has subsided, relapses may occur (Weaver, 1941), which simulate the picture of the subacute form. An unusually prolonged course is often complicated by other factors, especially neoplastic diseases.

**Pathology.** Pullet disease is characterized morphologically by dehydration, necrosis of the liver (spottiness) and pancreas (chalkiness), hemorrhages on the serous membranes, increased mucus in the intestine, various renal changes, and degenerations in skeletal muscle (fish flesh-like) and ovary (soft or broken follicles). The acute form exhibits congestive phenomena, or liver and muscle lesions, while renal changes predominate in the subacute form. Different combinations of such organic alterations in either gross or microscopic intensity produce a highly variable pathologic picture, especially as revealed by ordinary necropsy technique. This variability holds true for initial as well as follow-up specimens from the same outbreak, so that microscopic, hematologic, and chemical studies are necessary for complete diagnosis.

Birds affected with avian monocytosis are usually well developed and in good flesh, with a tendency to obesity. The appendages of the head appear congested, as well as the mucous membrane of the nasal passages. The vent feathers are soiled by urinary material. The *skeletal muscles,* especially the breast muscles, appear dehydrated, and show capillary injection. In some cases circumscribed pale, often turgent (fish flesh-like) areas are seen, which

microscopically represent patches of muscular degeneration: the myofibers are either in a state of granular disintegration separated by interstitial edema or, more characteristically, show loss of striation, fragmentation, and hyaline swelling, associated with mild polynuclear infiltration and incipient regeneration. In other words, they show the features of Zenker's degeneration, as seen in human typhoid fever and other toxic conditions (Fig. 29.1).

Although the *liver* may appear fatty or congested, an infrequent but most typical alteration in this organ is an evenly spaced studding with round



Fig. 29.1. Avian monocytosis. Section of breast muscle showing Zenker's degeneration. ×150.

yellowish areas about 1 mm. in diameter, which often have a minute hemorrhagic center. These foci may be few in number and may be associated with subcapsular petechiae. There is ordinarily no evidence of hepatic tumefaction or fibrinous exudation. Microscopically, the areas vary in size and represent typical focal necrosis of no particular zonal orientation (Fig. 29.2) ; pathogenetically they seem to develop either on the basis of simple coagulative necrosis of hepatic cells, or the accumulation of hyaline material in the Kupffer cells leading to sinusoidal thrombosis. The necrotic foci often undergo secondary polynuclear infiltration and may later be replaced by regenerating liver cells. The rest of the parenchyma shows marked biliary stasis, especially in the larger ducts (Fig. 29.3).

The *serous surfaces* often reveal multiple but comparatively few and widely spaced punctiform hemorrhages. These tend to occur on the visceral surface of the sternum, on the gizzard and abdominal fat, and on the pericardium. Microscopically, the peritoneal surface of the visceral organs is

frequently seen to be covered by a homogeneous eosinophilic material which is infiltrated with heterophils and spherical eosinophilic globules (Fig. 29.4) which are apparently derived from broken egg cells (Jungherr and Levine, 1941).

The *spleen*, as a rule, presents a normal appearance. The lack of tume-
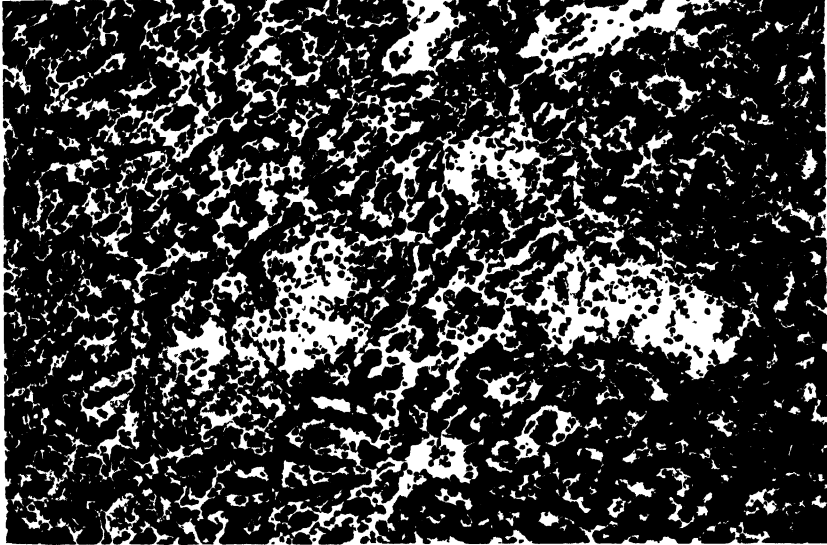


FIG. 29.2. Avian monocytosis. Section of liver showing focal necrosis. ×150.
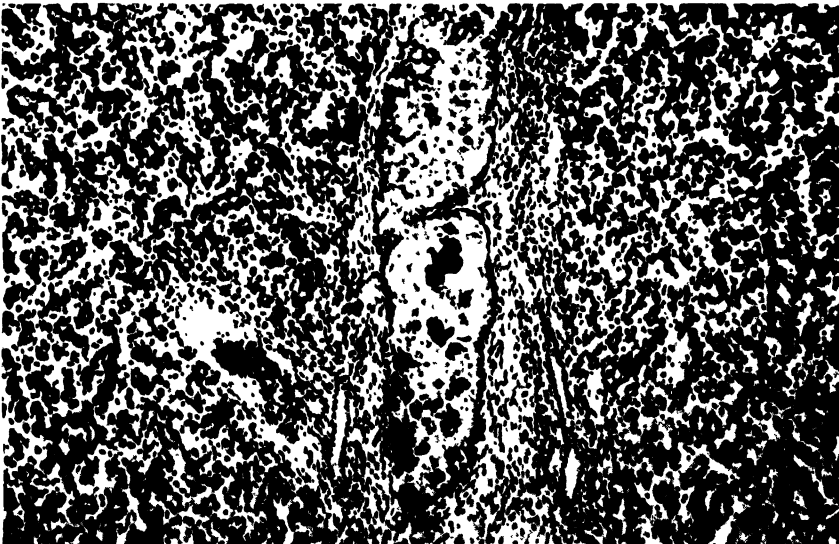


FIG. 29.3. Avian monocytosis. Section of liver. Severe bile stasis. ×150.

faction is helpful in the differential diagnosis and elimination of bacterial and leukotic diseases. Small necrotic foci are observed at times, together with bile- and hemosiderin-laden phagocytes. The *pancreas,* which normally displays a pinkish-gray homogeneous color, is apt to present a chalky appearance which resolves itself into numerous fine whitish areas on close inspection. This change, according to microscopic observation, seems to be brought about principally by cloudy swelling in the center of the acinar lobules, a process which may go on to karyorrhectic necrosis (Jungherr and Matterson,
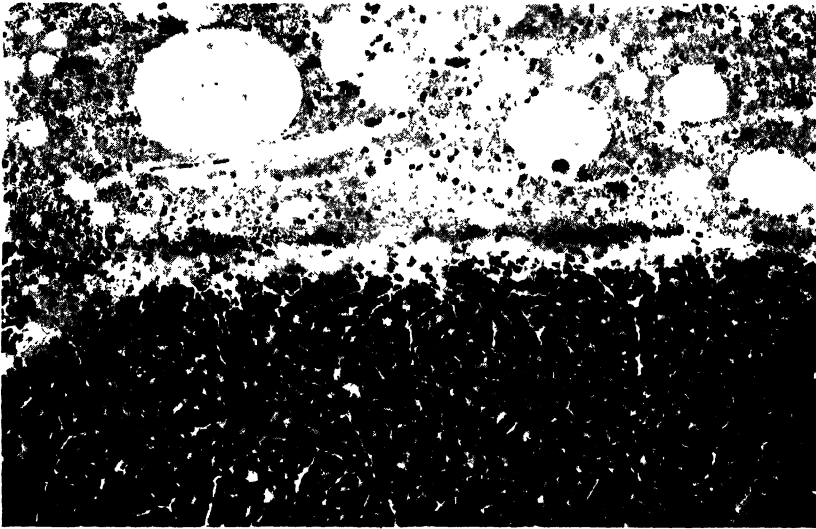


FIG. 29.4. Avian monocytosis. Section of pancreas. Serosa shows eosinophilic exudate with globules derived from egg cells. ×150.

1944). In addition, the size of the Langerhans islets appears sometimes increased; their cells are swollen or show here and there pale eosinophilic intranuclear inclusions, which may represent colloidal degeneration products (Jungherr and Levine, 1941). Similar inclusion bodies have since been observed in chickens not known to be affected with avian monocytosis by Lucas (1947), which circumstance would cast doubt upon their specific pathologic relation to the disease under discussion.

The external surface of the *intestine* is unaltered. The lumen of the ileum is usually filled with turbid tenacious mucus which is removable as a perfect cast. Histologically the changes are those of catarrhal enteritis. There may be desquamation of the epithelium with the subepithelial zones showing marked increase in cellularity. The inflammatory cells are composed chiefly of mononuclears, lymphocytes, and histiocytes. It is not uncommon to find many cystic crypts containing inspissated mucus (Jungherr and Matterson, 1944).

The gross lesions of the *kidneys* present a gradient from insignificant changes to marked enlargement, especially of the anterior lobes, and finally the familiar picture of uric nephritis known as visceral gout. Microscopic alterations are frequent in grossly "normal" kidney tissue; they are often of patchy distribution and vary in character. In the most acute cases one sees extensive cloudy swelling, pyknosis and desquamation of the epithelium of the proximal convoluted tubuli, a point which can be evaluated safely only in fresh autopsy material. Other definite renal changes consist of dilatation
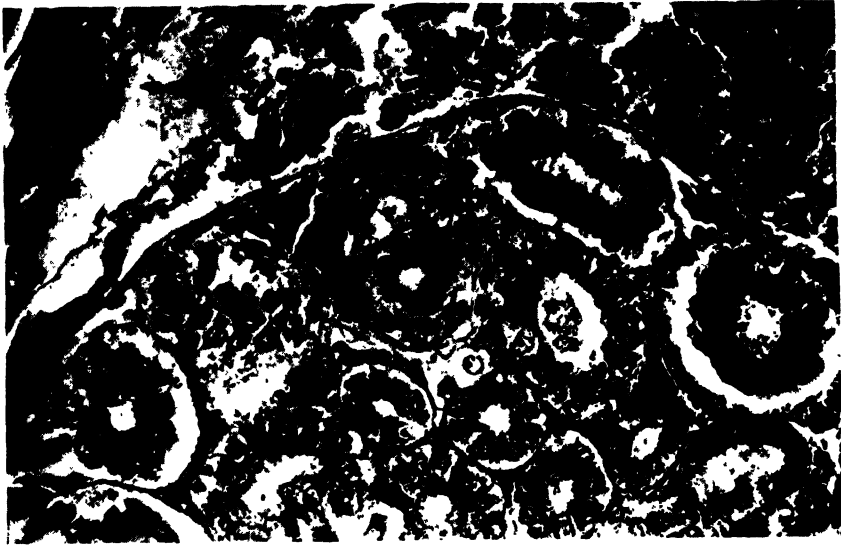


FIG. 29.5. Avian monocytosis. Section of kidney showing a large cast in the center. ×500.

of tubuli associated with flattening of the epithelium and formation of hyaline casts (Fig. 29.5) and pseudo-giant cells (Fig. 29.6) from infolding epithelium. The larger of these foci may show crystalloid radiating centers considered to be pathognomonic for uric nephritis. In protracted cases the tubuli show many cellular casts composed of disintegrating heterophils. The glomeruli likewise may exhibit significant alterations in avian monocytosis, such as thickening of the basement membrane, protein precipitate in Bowman's space (Fig. 29.7), adhesions, and dilatation (loculation) or hyaline thrombosis (Weaver, 1941) of the tuft capillaries. Fibrous obliteration of the glomeruli does occur in some instances.

The *ovary*, often being in full production, quite commonly presents irregular soft or broken egg follicles. The yolk material is of normal consistency. Massive fibrinous exudate around the follicles is not characteristic, and if present is probably due to secondary bacterial changes. The microscopic appearance of broken egg yolk material on the serous membranes has been described.

**Hematology.** The blood may show severe hemoconcentration, increased viscosity and coagulability, and low venous pressure. For these reasons it is sometimes difficult to obtain good blood samples by venepuncture. Birds are apt to die in the process. The hemoconcentration is reflected in increased hemoglobin values averaging 15.1 grams per cent in severely affected birds (Jungherr and Matterson, 1944).

In hematologic studies by the above authors, which have since been extended to over 100 cases, there was a consistent but moderate leukocytosis,
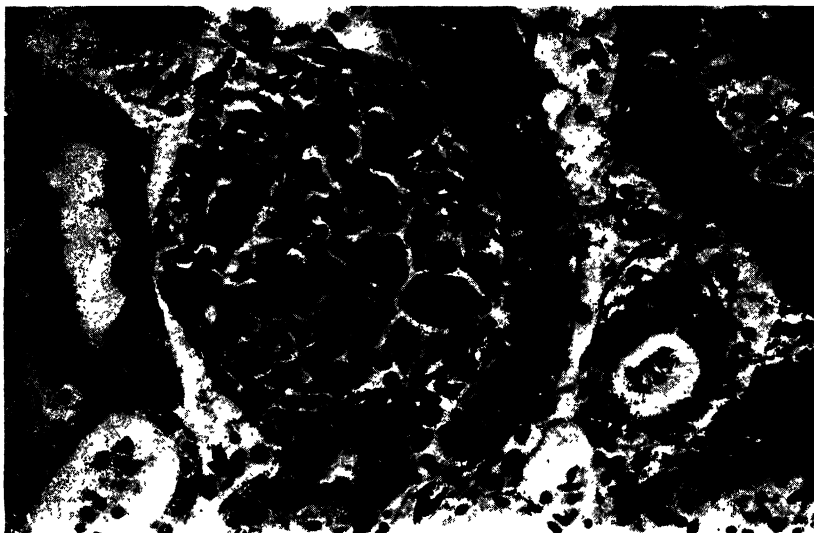


FIG. 29.6. Avian monocytosis. Section of kidney showing pseudo-giant cells in tubuli. ×500.

averaging 40,000 per mm.[3] The most significant change in the blood picture was a relative and absolute monocytosis which averaged about 20 per cent or 8,000 per mm.[3], respectively, in comparison with the normal of 8.9 per cent for females or 1,700, according to Olson (1943). The intensity of the blood changes varied with the clinico-pathologic picture and seemed to be particularly marked in cases of kidney involvement. As a rule, the majority of the birds in a specimen consignment showed the monocytic shift which sometimes constituted the only morphologic evidence of the disease. In stained smears the monocytes were ordinarily of the large, mature type and were sometimes characterized by basophilic cytoplasm and rounded nuclei suggestive of immaturity. Mitotic figures were rare.

The significance of the hematologic findings increased with the certainty with which other diseases such as fowl typhoid and fowl cholera could be ruled out. Since the blood changes seemed to represent the outstanding common denominator in pullet disease cases of varying intensity, the scien-

tific term "avian monocytosis" was proposed (Jungherr and Matterson, 1944).

**Chemical pathology.** Clinical resemblance of avian monocytosis to uremia and the pathologic evidence of kidney involvement and eclampsia-like hepatic necrosis emphasize the importance of the chemico-pathologic aspects. The earlier studies (Jungherr and Levine, 1941) have been extended by Levine and Jungherr (1941). It appears now that the blood of birds affected with pullet-disease-like conditions shows high average values for
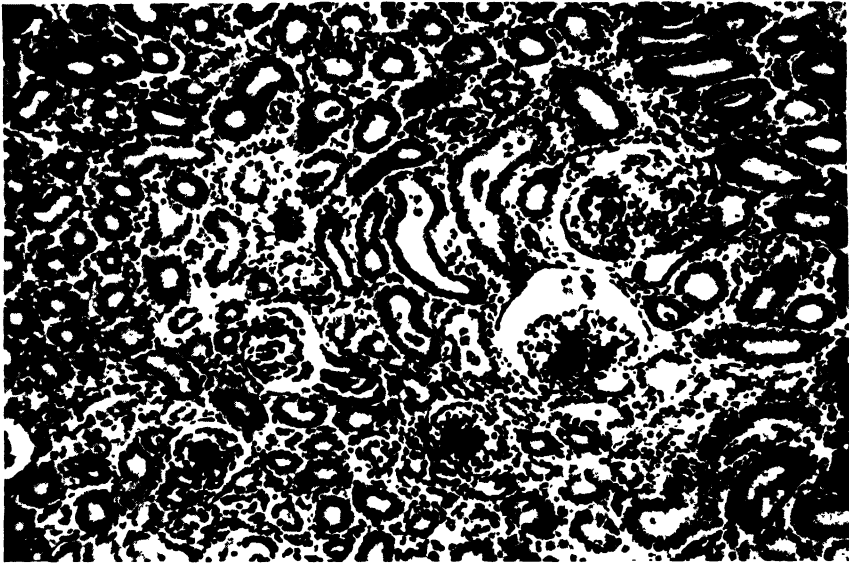


FIG. 29.7. Avian monocytosis. Section of kidney showing dilated glomerular space containing desquamated cells. ×150.

nonprotein nitrogen (26.8 mgm. per cent), and especially for uric acid (18.9), approximately normal values for phosphorus (6.5), magnesium (2.36), and total ketone bodies (15.5), and usually low values for calcium (13.9 mgm. per cent). The average value for glucose (200) is somewhat high, but wide variations are encountered in both affected and normal birds. In severe cases of avian monocytosis, the values for serum potassium are slightly below normal while those for whole blood potassium are high. Total chlorides may be strikingly low (Jungherr and Matterson, 1944). This chemico-pathologic picture is in keeping with a uremic concept of pullet disease.

Very acute cases of avian monocytosis usually fail to show high values for uric acid, but the nonprotein nitrogen may be increased. The difficulties connected with getting a representative number of such cases for examination have been pointed out.

Urine analysis of birds with high blood uric acid usually reveals a glycosuria; this may be due to the hyperglycemia, defective reabsorption on the part of damaged renal tubuli, or both. A reducing substance in birds suffering from experimental nephritis was first observed by Dworin, Jungherr, and Cook (1941) in this laboratory, and confirmed and identified in field cases of pullet disease by Levine and Jungherr (1941). Albuminuria was frequently observed in laying birds when the urine was obtained by the cloacal technique of Davis (1927), and Coulson and Hughes (1931), while urine obtained by cannulization of exteriorized ureters (Hester *et al.*, 1939–40) was free from albumin. Thus, the albumin seemed to be due to admixture of secretions from the genital tract, and to be of no pathologic significance.

**Differential diagnosis.** There are few examples in poultry pathology where differential diagnosis is of greater importance than in avian monocytosis. Some phases of the clinical and to a certain extent the pathologic picture of this disorder can be brought about by any of the common infectious diseases such as fowl cholera, pullorum disease, and fowl typhoid. For this reason it is of diagnostic significance to exhaust the possibilities of demonstrating a specific bacterial agent by cultural, serologic, and animal inoculation tests. Sporadic cases of pullet disease have some points in common with internal forms of the avian leukosis complex, which must be excluded on the basis of histologic and hematologic studies.

Associated conditions such as internal parasitic and protozoan diseases, bacterial and fungous diseases, and respiratory diseases, may obscure the syndrome under discussion. In sporadic cases it may be difficult to ascertain the primary condition and to differentiate between cause and effect. Flock outbreaks can often be suspected from the anamnestic data.

**Etiology.** The exact factors involved in the causation of the field syndrome of avian monocytosis are unknown. Etiologic studies have been concerned with the possibility of nephrotoxic substances particularly in wheat, physical factors such as overheating and dehydration, and of infectious agents.

Severe cases of avian monocytosis represent essentially a uremic condition referable to renal damage. Pathologic involvement of the liver in renal diseases is recognized in man under the term "liver death" and/or hepatorenal syndrome (Wilensky, 1939). Experimental nephrotoxicoses in birds, produced by a variety of factors, show a striking resemblance to avian monocytosis in clinical, chemical, and pathologic aspects. Thus, repeated intramuscular injections of potassium dichromate (0.001 per cent of body weight) caused uric nephritis together with occasional necrobiosis of liver, pancreas, and skeletal muscle (Jungherr and Levine, 1941). Feeding to chicks certain inorganic acids, particularly sodium citrate and acetate, pro-

duced the so-called "salt effect" which was preventable by potassium salts (Correll, 1941). In confirmation of this work, Scott, Jungherr, and Matterson (1944) found the salt effect to be indistinguishable from visceral gout or uric nephritis and to be preventable by potassium-rich molasses and potassium chloride. The latter substance also seemed to have a certain curative effect on spontaneous avian monocytosis. Selye produced nephrosclerosis in chicks by repeated subcutaneous injection of desoxycorticosterone acetate (1942), by watering with physiologic salt solutions or both (Selye and Stone, 1943), and believed the experimental condition to resemble avian monocytosis (1943). High protein diets alone, although producing articular gout in turkeys (Bollman and Schlotthauer, 1936b) and in chickens (Oppenheimer, 1941; Oppenheimer and Kunkel, 1943) apparently had no such damaging effect on the kidneys.

Investigating the popular claim that avian monocytosis represented a form of wheat poisoning, Quigley subjected it to experimental inquiry (1943) and obtained some epizootiologic as well as experimental support for this belief (1944a). He failed to find chemical differences between pullet disease-inducing and noninducing wheat samples, but found the former to have lowered germination ability (1944b) and to be associated with a high bacterial-low fungal flora (Petty and Quigley, 1947).

This entire subject of possible causes of visceral gout in birds has been recently reviewed by Stonebrink (1947).

High atmospheric temperatures have long been considered as a possible factor, as indicated by the synonym "summer disease." Yeates et al. (1941) in their studies of the reactions of domestic fowl to hot atmospheres failed to observe pullet disease. Jungherr in cooperation with Scott and Matterson (1946) examined chickens which had been kept at constant high atmospheric temperatures with or without adequate water supply, but failed to find a condition which would fit the diagnostic criteria of avian monocytosis.

No bacterial organism has been found to be constantly associated with avian monocytosis, except for the unconfirmed report of Weisner (1941) that the disease is caused by certain strains of *Escherichia coli*.

Waller (1942, 1944a) reported the isolation of a filtrable agent from the blood, liver, feces, and eggs of birds affected with the acute form. The virus (1944b, 1945) could be cultivated on the chorio-allantoic membrane of 8- to, 9-day-old chicken embryos where it produced a compact or circular lesion with radiating processes, stunting of the embryo and death in about 12 per cent. Turkey and duck embryos usually succumbed to the inoculation. Injection or feeding of the freshly isolated virus caused a nonfatal sickness (in about 50 per cent) characterized by subcutaneous edema, widespread petechiation, tumefaction of the parenchymatous organs, and catarrhal enter-

itis. The experimental disease in chickens and turkeys was accompanied by marked heterophil leukocytosis which reached its peak about 96 hours post-inoculation.

Blood sera from birds that had recovered from the experimental or spontaneous disease were capable of agglutinating washed killed *Salmonella pullorum* organisms which had been allowed to adsorb virus from infected allantoic fluid.

Based on the refractivity of birds to reinoculation, a live vaccine was prepared from infected chorio-allantoic membranes dried in vacuo over anhydrous calcium chloride and used on about 44,000 birds with encouraging results.

In evaluating the infectious and noninfectious etiologic theories of avian monocytosis, it should be kept in mind that so far the isolation of a filtrable agent has not been confirmed in the literature (Jungherr and Matterson, 1944). Failure to do so may be entirely technical in nature but demonstrates the difficulties involved, particularly for diagnostic purposes. Furthermore, known affected birds or their droppings have been brought in contact with presumably susceptible birds without evidence of spreading the disease. Waller (1944b) rightly pointed out the unlikeliness of many birds being attacked simultaneously by uremia of noninfectious origin. On the other hand, to explain the often explosive outbreaks of avian monocytosis on an infectious basis, one would have to assume that the viral agent is already widely seeded in the susceptible population, and that its pathogenic action is set into motion by secondary nonspecific factors (Jungherr and Matterson, 1944).

**Treatment and control.** Certain therapeutic measures have been used in the field, usually with favorable reponse if applied in the early stages of the acute form of the disease. However, it is often difficult to say how much of the apparent flock improvement can be credited to the treatment.

In general, the measures consist in providing an abundance of clean, readily available water, and cool, well-ventilated quarters, and in reducing consumption of grain. If possible, birds should be allowed access to runways or shaded range. Flushes are contraindicated, in view of the severe dehydration.

Some observers claim to have obtained good results from the use of 1:2,000 copper sulfate or 1:4,000 potassium dichromate in the drinking water (Weisner, 1941). Molasses has been used widely and has some justification because in high doses it prevents experimental nephrotoxicoses probably on account of its potassium content (Scott, Jungherr, and Matterson, 1944). Molasses may be used either in the drinking water (2 per cent) or administered in a mash composed of equal parts of bran and rolled oats, mixed with 10 to 30 per cent of molasses and water in amounts sufficient to

obtain a crumbly consistency. The treated mash is given on alternate days for a period of 3 hours, after withholding food for about 2 hours.

Instead of molasses potassium chloride or good fertilizer grade of muriate of potash (containing at least 60 per cent $K_2O$) may be used at the rate of 0.5 per cent in the water for the first 7 days, and if necessary, at the rate of $1\frac{1}{2}$ per cent in the feed for an additional 7 days. According to Van Ness (1947a, 1947b), under experimental conditions, prolonged or excessive use of potassium chloride may have untoward effects which have not been observed in the field.

If the virus-etiology of avian monocytosis finds confirmation by further demonstration of the causative agent, specific vaccination may be contemplated in recurrent outbreaks. At the present time, the specic vaccine is not available commercially.

Control measures should be attempted along the lines of good management. Routine prophylactic vaccinations (fowl pox, etc.) are best carried out during the early growing period, preferably at the age of two months. At the time of housing, the birds should be well fleshed, but not fat; transfer from the range to confinement must be made gradually with a minimum of disturbance to the birds. Shade, ventilation, and water supply are important factors, and mash feeding should be intermittent during the critical period.

## REFERENCES

Beaudette, F. R.: 1929. X disease. Poultry Path. Notes. N. J. Agr. Exper. Sta. 1:6–7.

———: 1940. In "Poultry information-please." Proc. 44th Ann. Meeting U. S. Livestock Sanitary Assn., p. 137.

Bollman, J. L., and Schlotthauer, C. F.: 1936a. Uremia in turkeys. Jour. Am. Vet. Med. Assn. 99:313.

——— and Schlotthauer, C. F.: 1936b. Experimental gout in turkeys. Am. Jour. Digestive Dis. and Nutr. 3:483.

Bullis, K. L.: 1940. Unknown disease. Proc. 13th Ann. Conf. Lab. Work. in Pullorum Disease Control. Mass. Agr. Exper. Sta., Mimeo. Rep.

Correll, J. T.: 1941. The biologic response of chickens to certain organic acids and salts with particular reference to their effect on ossification. Jour. Nutr. 21:515.

Coulson, J., and Hughes, J. S.: 1931. Collection and analysis of chicken urine. Poultry Sci. 10:53.

Davis, R. E.: 1927. The nitrogenous constituents of hen urine. Jour. Biol. Chem. 74:509.

Dworin, M., Jungherr, E., and Cook, W. B.: 1941. Unpublished data.

Gordon, R. F., and Blaxland, J. D.: 1945. The occurrence in England of outbreaks of disease in poultry resembling the so-called pullet disease in America. Vet. Jour. 101:3.

Hester, H. R., Essex, H. E., and Mann, F. C.: 1939–40. Secretion of urine in the chicken (*Gallus domesticus*). Am. Jour. Phys. 128:592.

Hurt, L. M.: 1941. Pullet disease. Los Angeles County, Calif., Livestock Dept., Ann. Rep., pp. 52–53.

Jungherr, E.: 1945. Report of the committee on transmissible diseases of poultry. Proc. 49th Ann. Meet. U. S. Livestock Sanitary Assn., p. 65.

——— and Levine, J. M.: 1940. The pathologic concept of so-called "pullet disease." Poultry Sci. 19:354.

——— and Levine, J. M.: 1941. The pathology of so-called pullet disease. Am. Jour. Vet. Res. 2:261.

Jungherr, E., and Matterson, L. D.: 1944. Avian monocytosis, so-called pullet disease. Proc. 48th Ann. Meet. U. S. Livestock Sanitary Assn., p. 185.

Lucas, A. M.: 1946. Private communications and Eighth Annual Rept. of U. S. Regional Poultry Research Laboratory, East Lansing, Mich., pp. 9 and 10.

Olson, Jr., C.: 1943. Avian hematology. In H. E. Biester and L. DeVries, Diseases of Poultry, Iowa State College Press, Ames, Iowa. Pp. 67–84.

Oppenheimer, E. H.: 1941. The lowering of blood uric acid by uricase injections. Bul. Johns Hopkins Hosp. 58:190.

———, and Kunkel, H. G.: 1943. Further observations on the lowering of blood uric acid by uricase injections. Bul. Johns Hopkins Hosp. 73:40.

Petty, A. M., and Quigley, G. D.: 1947. The microflora of wheat feeds as related to the incidence of blue comb in chickens. Poultry Sci. 26:7.

Quigley, G. D.: 1943. Is blue comb of fowls produced by wheat? Poultry Sci. 22:267.

———: 1944a. The effect of wheat upon the incidence of pullet disease or blue comb. Poultry Sci. 23:386.

———: 1944b. Germination differences of wheat utilized in a study of pullet disease. Poultry Sci. 23:547.

Ryff, J. F., and Stafseth, H. J.: 1942. A cholera-like disease of poultry. Vet. Med. 37:294.

Scott, H. M., Jungherr, E., and Matterson, L. D.: 1944. Possible role of potassium in pullet disease. Proc. Soc. Exper. Biol. and Med. 57:7.

———, Matterson, L. D., and Jungherr, E.: 1946. Unpublished data.

Selye, H.: 1942. Production of nephrosclerosis by overdosage with desoxycorticosterone acetate. Canad. Med. Assn. Jour. 47:515.

———: 1943. Production of nephrosclerosis in the fowl by sodium chloride. Jour. Am. Vet. Med. Assn. 103:140.

——— and Stone, H.: 1943. Role of sodium chloride in production of nephrosclerosis by steroids. Proc. Soc. Exper. Biol. and Med. 52:190.

Spanner, R.: 1925. Der Pfortaderkreislauf in der Vogelniere. Gegenbaurs morphologisches Jahrb. 54:560.

Stonebrink, B.: 1947. De pathogenese van jicht bij vogels. Tijdschrift voor Diergeneesk. 72:164.

Van Ness, G.: 1947a. Potassium as an excitant to blue comb. Poultry Sci. 26:557.

———: 1947b. The production of so-called pullet disease. Poultry Sci. 26:304.

Waller, E. F.: 1942. Isolation of a filtrable virus from chickens affected with blue comb disease. Science 95:560.

———: 1944a. Virus etiology of blue comb disease. Proc. Sixteenth Ann. Conf. Lab. Work. in Pullorum Disease Control. Univ. Conn. Coll. Agr., Mimeo. Rep.

———: 1944b. Blue comb disease. Proc. 48th Ann. Meet. U. S. Livestock Sanitary Assn., p. 171.

———: 1945. Blue comb disease. N. H. Agr. Exper. Sta., Tech. Bul. 85:3.

———, Tepper, A. E., Halpin, R. B., and Davis, H. A.: 1942. The etiology, pathology, and prevention of contagious indigestion. N. H. Agr. Exper. Sta. Rep., Bul. 345:59.

Weaver, C. H.: 1941. Bright's disease in the avian subject. Proc. 14th Ann. Conf. Lab. Work. in Pullorum Disease Control, Federal Dept. Agr., Mimeo. Rep., Ottawa, Canada.

Weisner, E. S.: 1941. How the veterinarian can develop a poultry practice. Mich. St. Coll. Vet. 1:10.

Wilensky, A. O.: 1939. Occurrence, distribution, and pathogenesis of so-called liver death and/or the hepatorenal syndrome. Arch. Surgery 38:625.

Yeates, N. T. M., Lee, D. H. K., and Hines, H. J. G.: 1941. Reactions of domestic fowls to hot atmospheres. Proc. Roy. Soc. Queensland 53:105.

CHAPTER THIRTY

## NEOPLASTIC DISEASES OF THE CHICKEN

By WILLIAM H. FELDMAN, *Division of Experimental Medicine,*
*Mayo Foundation, Rochester, Minnesota*

*and*

CARL OLSON, JR., *Department of Animal Pathology and Hygiene,*
*University of Nebraska, Lincoln, Nebraska*

☆    ☆    ☆

Neoplastic diseases are relatively common in the domestic chicken. During the past fifteen years those concerned with the health of poultry have become aware of the serious loss to the industry due to these conditions. This awareness has awakened and stimulated interest in a group of diseases that have not always received the attention their importance merits. A vast amount of information on neoplasia has been gathered from the study of tumors of the various species of animals, and some of this information may be applied to the disease as it affects chickens. Much of our present knowledge of neoplasia has been gained in relatively recent times, and additional information is being supplied constantly from the results of research. Such new information must be correlated and when possible integrated with the older facts. Obviously, this may lead to entirely new conceptions, and a consideration of tumors must bear this possibility in mind.

An attempt is made in the following presentation to describe briefly the various forms of neoplastic disease that occur in the chicken and to discuss their salient characteristics. This contribution is intended to supply the information necessary for a pathologist to make a differential diagnosis of neoplasia in the chicken, provided the case in question is one of the commoner forms of tumor. Key references to the literature are provided for those who wish to seek more detailed information.

The cases of tumor which form the basis for discussion are from the combined collections of the authors. These are supplemented in some instances by cases described in the literature. Since the material studied was obtained from several widely separated geographic regions, it may be said in general that the descriptions contained in the text that follows are fairly representative of neoplasia of chickens as encountered in this country.

## INCIDENCE

Although neoplastic disease is generally recognized as one of the more common diseases of the domestic chicken, its incidence can be estimated in only a general manner. Schneider (1926) studied the records of the Storrs Egg Laying Contest, the Harper Adams Trials, and a farm flock which included about 11,000 birds. She concluded that the usual incidence of tumors in chickens six to eighteen months old was about 2 to 3 per cent. Curtis (1915) stated as the result of a survey made in Maine that the incidence of tumors was 8.98 per cent. This figure was based on the finding of seventy-nine cases of tumor diagnosed on gross examination at the time of necropsy of 880 birds during an eight-year period. The early reports of Curtis and Schneider provide only general information since histologic studies of their cases were not made. Other reports of a similar nature are available from various sources such as the diagnostic services maintained by various institutions. Such data add but little to our knowledge of the incidence of neoplasia in the chicken, for usually diagnoses are made without microscopic study, and the terminology and classifications are likely to be misleading.

European workers have compiled data bearing on the question. Their data consist largely of material collected over a period of years in diagnostic laboratories and usually are substantiated by histologic study. Hoogland (1929) reported from Holland that in the period from 1906 to 1928, 1,707 birds were examined, and 176 tumors were found, an incidence of about 10 per cent. A larger survey was reported by Eber and Malke (1932) from the University of Leipzig covering a period of thirty-two years. In this period 11,903 chickens were examined, and 371 cases of neoplasia were found, an incidence of 3.12 per cent. Babic (1931) found forty-two tumors among 647 chickens examined at Zagreb, Yugoslavia, during the interval between 1923 and 1931. Olson and Bullis (1942) made a survey of the material received by the diagnostic laboratory at Massachusetts State College during a two-year period. A total of 2,304 chickens more than six weeks of age were examined, and 297, or 12.9 per cent, were affected with neoplasia. Campbell (1945) found 386 (18.7 per cent) cases of neoplasia among 2,063 chickens examined over a five-year period at Edinburgh, Scotland.

A survey such as that made by Goss (1940b) on six flocks of chickens having a total population of more than 24,000 birds provides useful information. About 6 per cent (1,445) were found to have tumors at necropsy. Actually, 7,408 birds were examined at necropsy, which provides a figure of 19.51 per cent incidence of tumor in birds on which necropsy was performed that may be compared with the figures of Curtis, Hoogland, Eber, and Malke, and Olson and Bullis. Five of the flocks surveyed by Goss were under observation for a year; the sixth was under ovservation for only three months.

Ask-Upmark (1938) reported what he believed to be an epidemic of tumors in a flock of chickens in Sweden. Between twenty and twenty-five birds from a flock of 100 died within a year. Only five of the birds were actually examined; yet all five were affected with carcinoma of the ovary with extension to the viscera. Olson (1942) studied the tumors that occurred in a small poultry flock in which the incidence of disease was unusually high. The flock numbered forty-eight birds at the beginning of the study, and at the time the last bird was slaughtered, three years later, a total of thirteen birds (27 per cent) had died with neoplastic disease. More recently a flock of 465 birds has been studied, and in a two-month period twenty-nine, or 6.23 per cent, were found affected with lymphocytoma. Such instances are no doubt unusual; however, they serve to illustrate the difficulty of arriving at a figure which may be said to represent even the general incidence of neoplasia in chickens.

Various factors obviously will affect the result of any survey on the incidence of neoplasia. Some of the more important factors are age of the group under survey, length of period covered by the survey, the genetic composition or inheritable tendencies of the group under survey, and certain other factors as yet unknown which may cause a high incidence of certain types of tumors. As these factors may not be comparable among different populations of chickens, the incidence of neoplasia will be variable and unpredictable. Further research is needed on the general incidence of neoplasia, for the results of such surveys may be most enlightening as to factors which influence the incidence of specific types of tumors.

In this connection a plea may be made for thoroughness in such work. Each case of neoplasia should be studied carefully. Both the gross and histologic aspects of the material should be recorded, and the type of neoplasia should then be classified properly. This type of work should be done by a pathologist trained especially for the task. The practice of considering all cases of neoplastic disease simply as "tumors" and of diagnosing specific forms of neoplasia by hasty examination of gross specimens has been perhaps justifiable in the past. However, rapid progress toward the solution of the problems caused by the more common tumors of chickens will depend on more accurate and sound research work.

Much time and effort are being devoted to these problems by many workers. The results of these investigators must have a common basis for correlation, which is an accurate diagnosis and common terminology of the conditions that they encounter. These comments pertain to information collected as a part of a research project. The situation is different for one whose only concern is the distinction of neoplastic disease from a granulomatous process. This would be true in the case of laboratories established for the purpose of providing routine diagnoses on material submitted from com-

mercial poultry flocks. In this instance a careful study of the gross specimen and elimination of pathogenic bacteria as a cause of the lesion may enable the pathologist to arrive at a presumptive diagnosis without the aid of a histologic examination. Many cases of neoplasia will be sufficiently typical to permit their identification from gross examination alone, providing that the observer is experienced and has a sound understanding of the fundamental histology of neoplasia. Reports of diagnoses based only on gross examination of the specimens should be so indicated for the information of the reader.

Some forms of neoplasia are much more common than others. Lymphocytoma is recognized generally as the most common tumor found in chickens. It is so common that the mere mention of tumors of chickens is likely to conjure, in the minds of many, thoughts of lymphocytoma to the exclusion of the many other varieties of neoplasia that also occur in chickens. Collectively, the other varieties cause much loss, and other types of tumors may be a serious problem in some flocks of poultry.

Scientific interest was aroused more than thirty years ago in two types of neoplasia of the chicken which were found to be transmissible by means of an ultramicroscopic agent separable from tumor cells. These tumors were fowl leukosis and certain forms of tumors of connective tissue. They have been subjected to intensive study, and a large volume of information concerning their behavior in experimental chickens is available. Despite the knowledge gained from studies of the experimentally transmissible tumors of the chicken, we have no evidence or even conclusive indication of the mode of spread of these diseases from one bird to another under natural conditions.

## CLASSIFICATION OF NEOPLASMS OF CHICKENS

Many schemes or systems have been proposed by pathologists for the systematic grouping of true tumors. Some schemes are extremely simple, such as one that provides for only two groups of tumors, one benign and the other malignant. Other classifications are more elaborate, and some are indeed complicated and even confusing. A fairly satisfactory system of classifying tumors is that which depends on the type cell from which the tumor originates. Such a classification is based on the embryogenesis and histologic identity of the respective cells that make up normal tissues and takes cognizance of the fact that tumors may arise from any of the distinct varieties of cells that are concerned normally in the structural or functional welfare of the body. One practical difficulty with this system of classification is that although cells of normal or mature tissues usually possess characteristics by which they can be recognized readily, unripe or immature cells such as make up rapidly growing tumors, or those cells representing primitive tissues, may exhibit so little differentiation that recognition of the type cell may be difficult or impossible.

The classification shown in Table 1 is not original and has certain short-comings. However, it does provide, we belive, a fairly simple and practical outline by which most tumors that arise in poultry may be grouped. While it may be sufficient for most utilitarian purposes to know if a given tumor is benign or malignant, for the sake of certain clinical and histologic considerations it is frequently desirable to use terms that are more specific. These are provided in Table 1.

TABLE 1
CLASSIFICATION OF NEOPLASMS OF CHICKENS *

I. Tumors of connective tissues

   A. Benign
      Fibroma
      Myxoma
      Lipoma
      Osteoma
      Chondroma

   B. Malignant
      Fibrosarcoma
      Myxosarcoma
      Liposarcoma
      Osteogenic sarcoma (osteochon-
        drosarcoma)
      Chondrosarcoma
      Histiocytic sarcoma

   C. Special forms of connective
     tissue tumors
      Neurogenic sarcoma

II. Tumors of muscle tissue
      Leiomyoma
      Rhabdomyoma

III. Tumors of blood and lymph channels
      Hemangioma
      Lymphangioma

IV. Tumors of hemoblastic origin
      Lymphocytoma
      Myelocytoma
      Leukosis

V. Pigmented tumors
      Melanoma

VI. Tumors of nerve tissue
      Glioma
      Neuroblastoma
      Retinoblastoma
      Ganglioneuroma

VII. Tumors of epithelial tissues

   A. Benign
      Papilloma
      Adenoma

   B. Malignant
      Carcinoma

   C. Special forms of epithelial tumors
      Hypernephroma
      Arrhenoblastoma
      Dysgerminoma
      Granulosa-cell tumor

VIII. Tumors of serous membranes
      Mesothelioma

IX. Mixed tumors
      Thymoma
      Carcinosarcoma
      Teratoma
      Dermoid cysts
      Embryonal nephroma

* While some of the varieties of neoplasia listed have not been reported as yet in chickens, the fact that their occurrence is theoretically possible justifies their inclusion in this scheme of classification.

## TUMORS OF CONNECTIVE TISSUE

The widespread distribution of the different connective tissues throughout the body provides potential sources for a variety of connective tissue tumors. These tumors may be defined as benign or malignant neoplasms derived from certain mesenchymal elements such as ordinarily produce fibrous connective tissue, mucous connective tissue, cartilage, bone, and fat. Another potential source of connective tissue tumors is the free histiocyte which occurs normally associated with all loose connective tissues.

Tumors derived from the various connective tissues may be extremely simple in their constituents and easily recognized, or they may be more or less complex in structure and difficult to classify with certainty. Many variations of structure occur. Single benign fibroblastic tumors may be associated with edema or mucinous substances, and it may be difficult in the latter instance to determine whether or not one is dealing with a simple fibroblastic entity associated with mucinous degeneration or with a tumor that is primarily myxomatous. The connective tissue tumors composed of cartilage, fat, or bone are ordinarily not difficult to recognize if sufficient differentiation has occurred. However, in instances in which the type cells are more primitive and differentiation characteristic of adult tissue is absent or obscure, practical difficulties in the recognition of these tumors may be encountered.

*Frequency of occurrence.* Generally speaking, the benign connective tissue tumors are among the rarer neoplasms of chickens. In a series of 113 chicken tumors (exclusive of leukotic tumors) reported by Goss (1940a), only one benign connective tissue tumor—a fibroma—was listed. Among 237 tumors of chickens examined histologically by Eber and Malke (1932), eleven benign connective tissue tumors were found. These included one myxoma, five fibromas, and five lipomas. Hoogland (1929), of Utrecht, reported twelve fibromas among 176 chicken neoplasms. No other forms of benign connective tissue tumors were listed by Hoogland. Eight of Goss's series of 113 tumors were fibrosarcomas. In our collection malignant forms of connective tissue tumors are more frequent than the benign entities. In Jackson's (1936a) series of 203 neoplasms (including lymphocytoma and leukosis) of chickens, twenty-four malignant connective tissue tumors are listed. The different varieties were as follows: seven fibrosarcomas, one myxosarcoma, one osteochondrosarcoma, and fifteen histiocytic sarcomas. In the series of 384 neoplasms of chickens, inclusive of leukosis, presented by Olson and Bullis (1942), there were sixteen fibrosarcomas, three histiocytic sarcomas, five neurogenic sarcomas, one osteochondrosarcoma, and one fibrochondrosarcoma.

As in most other forms of neoplasia of the chicken, adequate precise statistical data on the incidence of connective tissue tumors are not available.[1] Our observations and the impressions obtained from the reports of others suggest that the benign connective tissue tumors occur infrequently and that the malignant varieties are at least of moderate frequency.

*Sites of occurrence.* As may be inferred, tumors of connective tissue origin may arise from any situation where the prerequisite parent cells occur. Fibromas, myxomas, and lipomas are most likely to arise from the integument, while simple tumors composed of cartilage or bone or a mixture of

---

· [1] In the older literature the term "sarcoma" was used in a rather nonspecific sense, and no attempt was made to separate the various types of sarcomas on the basis of the type cell.

these two tissues may be expected to arise from situations where cartilage or bone normally occurs. Certain multipotent mesenchymal cells may and sometimes do give rise to cartilage and bone in situations where these tissues ordinarily are not expected. Such multipotent cells account for certain so-called mixed tumors in which there is a combination of fibroblastic elements, cartilage, bone, and mucin. Fibrosarcomas have been observed in the integument, retroperitoneal region, the tissues of the abdominal wall, and the muscles of the breast and thigh. While occasionally fibrosarcomas arise in the interior of the body, the majority occur in the tissues of the exterior. Of the connective tissue tumors, the group designated histiocytic sarcoma is capable of the widest anatomic distribution. Situations in which this tumor has occurred include the wattle, esophagus, subcutis, liver, spleen, and ovary.

If there is a site of predilection for the occurrence of osteogenic sarcomas in chickens, it is not known. These tumors are among the less frequent neoplasms of chickens, and relatively few have been reported. They may arise wherever periosteal tissue occurs.

*Effects on the host.* Benign tumors of connective tissue origin ordinarily do not constitute a serious handicap to the well-being of the affected animal. However, they may affect the host adversely on account of their size and location. Those situated on the exterior of the body are subject to trauma and subsequent infection. Tumors situated in the nasal or the pharyngeal region may cause respiratory distress.

The malignant varieties of connective tissue tumors are potentially lethal, and the effect on the host depends on whether metastasis has occurred and what organs are affected. One gets the impression that these tumors, especially the fibrosarcomas and the histiocytic varieties, grow rapidly, and death or extreme debility may ensue relatively soon after the disease has become disseminated. Most of these tumors produce a progressive destructive disease that invariably results in death.

*Gross and microscopic description.* Like many other tumors those derived from connective tissue elements seldom have sufficient distinguishing gross characteristics to make their identification certain. However, gross differences do exist between those that are benign and those that are malignant. Fibromas are usually circumscribed, fleshy, nodular or oval masses that may be soft or firm (Fig. 30.1). A capsular covering usually can be recognized. Myxomas are likewise more or less circumscribed, of soft consistency, and the slimy mucinous or gelatinous character of most specimens is evident. A lipoma, being composed largely of fat, offers little difficulty in recognition. The identifying features of benign tumors of cartilage or bones should be obvious.

Gross features that might enable one to distinguish malignant connective tissue tumors from other malignant growths are not to be relied on when an

accurate diagnosis is desired. Although signs suggestive of malignancy, such as lack of encapsulation, multiplicity of lesions within the same general region, or the presence of contiguous implantations or distant metastatic growths, usually will permit of no doubt as to the malignancy of the process; specific features that might enable one to recognize the specific character of such a neoplasm are missing. The diagnosis of these tumors, as of most others, must depend on microscopic examination.

Since the gross appearance of these tumors is subject to wide variations dependent on their size and anatomic situation, it can be described only in
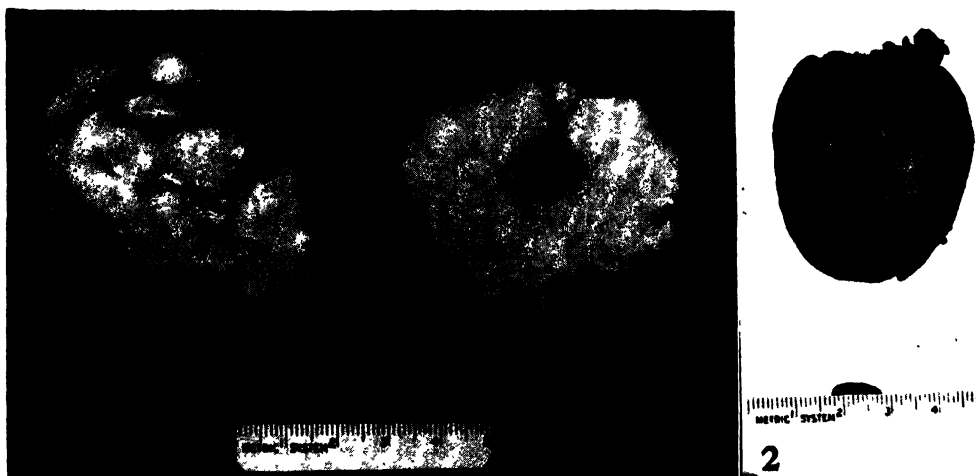


FIG. 30.1. 1—fibroma encircling the intestine. 2—fibroma in the ventricular wall of the heart.

general terms. Those on the exterior of the body are usually oval or hemispherical and flesh pink. Most of the large specimens on the exterior of the body erode the overlying skin and may present a granulated surface with brownish-black ulcerations due to trauma and subsequent hemorrhage.

Those of the interior of the body may consist of single or multiple fleshy masses of variable sizes. The surface is usually smooth, and if the tumor is in the ovary the contour may be and often is lobulated. If the tumor is primary in the ovary, transplantation to the contiguous situations usually results in the formation of innumerable small to large nodules of neoplastic tissue in the serosa of the intestines and mesentery. Fairly frequently the entire abdominal viscera become fused or matted together by the neoplastic tissue to such a degree as to make separation of the respective parts difficult.[2]

As mentioned previously, while these features are indicative of malignant

---

[2] Similar neoplastic involvement secondary to ovarian malignant lesions also occurs in carcinoma of the ovary and in lymphocytoma.

neoplasia, one should be cautious in concluding from the gross appearance that the condition represents a process of connective tissue derivation. Only a microscopic examination can supply the information necessary for a correct diagnosis.

*Special features.* One of the most interesting features of connective tissue tumors of the chicken is the transmissibility of some forms by filter-passing agents. The transmissibility of a considerable number has been studied, and a huge literature has accumulated which deals with these investigations. This feature cannot be covered adequately in a few paragraphs, and one interested should consult the excellent reviews of Claude and Murphy (1933) and Foulds (1934). A variety of such agents has been studied. They differ from each other both in the specific form of tumor they produce and in certain serologic characteristics. The existence of ultramicroscopic tumor-producing agents suggests the possibility of epidemics of tumors. However, evidence from experiments indicates the lack of contagiousness of such tumors. Carr (1944) reported no neoplastic disease in chicks hatched from hens that had recovered from the Rous No. 1 sarcoma. Yolk of eggs laid by the hens contained a substance which neutralized the action of the sarcoma agent. Previously Carr (1943) had demonstrated persistence of the neutralizing substances in blood serum of recovered fowls for one to two years which were believed due to latent virus still in the tissues of the hens. Duran-Reynals (1940) has reported similar neutralizing substances believed to exist as natural antibodies in the blood of adult normal chickens. An etiologic relation between certain fibrosarcomas and fowl leukosis is discussed in the section describing fowl leukosis. Some of the transmissible connective tissue tumors of chickens have been reproduced experimentally in other species of fowl. Perhaps most spontaneous connective tissue tumors of the chicken might prove transmissible if all the conditions conducive to success could be supplied. Duran-Reynals (1946a) was more successful with transplantation and demonstration of cell-free agents of sarcomas occurring in chickens five to ten months of age. Carcinogenic chemicals have produced connective tissue tumors in birds under proper experimental conditions. However, such tumors do not appear transmissible by cell-free agents (Murphy and Sturm, 1941a, 1941b; Peacock, 1946). Peacock (1946) studied the histology of fifteen transplantable, chemically induced sarcomas and three sarcomas that could be transmitted by cell-free agents. He states that slight but definitely recognizable differences exist between tumors induced by chemicals and those by cell-free agents.

**Fibroma and fibrosarcoma.** A fibroma in its simplest form consists of rather adult fibroblasts and narrow to wide strands of collagen disposed parallel to the longitudinal axis of the fibroblasts. The fibroblasts vary in size and number according to the rate of growth. Fibromas that progress

slowly show the greatest degree of differentiation in that more collagen is produced, and the resultant structure contains relatively few cells in proportion to the collagen present. The less differentiated forms are more cellular and less firm owing to a relatively small amount of collagen. The strands of collagen may be disposed in every direction, and regions in which the strands are arranged in an undulating fashion are seen frequently. Most fibromas of chickens are moderately to well supplied with large to small blood vessels, and we have observed channels suggestive of lymph vessels. In some instances markedly edematous regions occur in which the fluid causes separation and
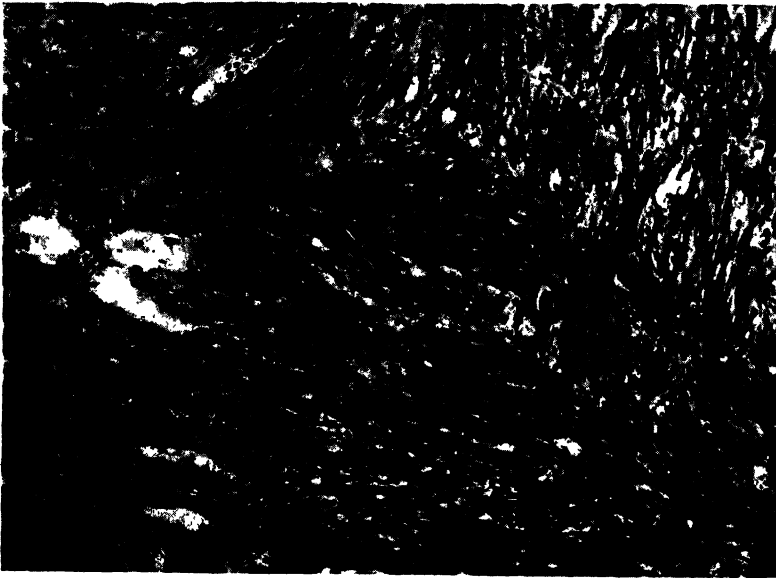


FIG. 30.2. Fibrosarcoma in the musculature of the breast. The neoplastic process was bilateral and had metastasized to the lungs. ×120.

retrogression of the collagen fibrils. The edema in these instances should not be confused with the mucinous product of myxomatous tumors. If infection has occurred because of erosions at the surface, various phases of necrosis may be recognized.

The essential features of fibrosarcomas are the immaturity of the type cells and the aggressive and destructive behavior of the neoplastic process (Fig. 30.2). Large, irregularly arranged, hyperchromatic fibroblasts are abundant, and mitosis is common. Collagen is present in variable amounts but is less abundant than in fibroma. Stromal elements, frequently disposed as irregular septa, may occur, and small blood channels usually can be recognized. In rapidly growing specimens, small to extensive regions of necrosis are likely

to occur. Occasionally, widespread edema is present, causing considerable separation of the fibroblastic components.

**Myxoma and myxosarcoma.** Myxomas consist of stellate or spindle-shaped cells surrounded by a homogeneous, slightly basophilic, mucinous matrix. Long cytoplasmic processes may extend from the stellate cells and become fused with the matrix and collagen fibrils. In the malignant form of myxoma the mucinous matrix is less abundant than in the benign form, and the type cells are proportionately more numerous and more immature. The histogenesis and structure of primary myxomatous tumors are closely related to those of the fibroblastic tumors, the essential difference being that in myxomatous tumors the fibroblastic cells are more specialized and are capable of producing mucin in addition to the usual products such as fibroglia, collagen, and elastic fibrils.

Our series contained three myxoblastic tumors. One occurred in a chicken, aged three to four years. The tumor arose in the subcutaneous tissues at the base of the right wing and extended to the humeroradial articulation. The exact genesis appeared to be in the lower zone of the corium. The process was highly invasive and destructive locally. Although the tumor had infiltrated into the adjacent muscle, strangely enough metastasis had not occurred. The second case was that of a nine-month-old pullet. The tumor was primary in the exterior of the ovary, and this organ was practically replaced by the neoplasm. The intestines, mesentery, peritoneum, and kidneys were involved extensively by direct extension from the parent growth. The infiltrative propensity of the neoplastic cells was illustrated by the fact that the tumorous process had penetrated through the muscle walls of the intestines in some instances. In the third case, that of a twenty-month-old hen, the tumor appeared to have originated from the ovary. Numerous implantations had occurred in the mesentery, especially where it joined the intestines. There were no signs of immaturity, and true metastasis had not occurred.

Olson and Bullis (1942) observed two cases of myxoma in chickens. In one case the tumor had occurred in a six-month-old pullet. It was attached to the left kidney and weighed 717 gm. It had remained localized. In the other case, that of a seven-month-old pullet, the tumor occupied the left orbital cavity and caused protrusion of the eye. Metastasis had not occurred.

**Lipoma.** Like other tumors of connective tissues, those derived from the lipoblast may be composed of adult cells and present the physical appearance of ordinary fat tissue, or the immature forms of the lipoblast may predominate, in which case the tumor presents a sarcomatous appearance. The microscopic appearance of lipoma is extremely simple. The process consists of compactly arranged, moderately large to extremely large polyhedral cells filled to capacity with a large fat globule or several small ones. The nucleus

may be crowded to the periphery of the cell and be entirely obscure. The stroma consists of narrow strands of connective tissue which provide support for the blood vessels. A thin capsular covering is usually evident. Grossly the yellowish, fatty consistency of lipomas of the chicken strongly suggests their true nature. Frozen sections stained with scarlet red provide convincing evidence of their lipomatous character.

**Osteoma and osteogenic sarcoma.** The rarity of these forms of connective tissue tumors among chickens has precluded the opportunity for their examination. Heim (1931) cited the report of one case of osteoma in the chicken. The brief descriptions which follow represent a study of tumors of bone from animals other than fowls. The structure of an osteoma simulates bone with the exception that much of the finer histologic detail is lacking. The process consists of a diffusely disposed acidophilic matrix of osseomucin separated at irregular intervals by collections of osteoblasts. Lamellae may be recognized, and the mimicry may include structures comparable to or suggestive of haversian canals. Osteogenic sarcomas are usually very cellular infiltrative growths which destroy the surrounding tissues and readily metastasize. The immaturity of the cells is usually evident, and mitotic figures are commonly numerous. The cells may be spindle-shaped, ovoid, or polyhedral, and foreign body giant cells occasionally occur. Although an osteogenic tumor is usually a highly cellular and frequently a rapidly growing tumor in which immature or undifferentiated cells may constitute the bulk of the structure, a few to many cells usually can be found in which sufficient differentiation has occurred to produce osseomucin. The finding of this substance, which has a homogeneous acidophilic appearance, is usually sufficient to reveal the true character of these tumors.

**Chondroma and chondrosarcoma.** Only a few cases of simple chondrogenic tumors in chickens have been reported (Heim, 1931). No specimens have been available from chickens for our study. It should be kept in mind that cartilage cells are the product of specialized fibroblasts, and in the early phases of their functional differentiation these cells have the appearance of ordinary mesenchymal cells. By a gradual process of transition adult cartilage cells finally are evolved which produce chondromucin in addition to collagen and elastic fibrils. The adult cartilage cell is the end phase of the transition process and is not capable of the production of other cartilage cells. These must arise from certain fibroblasts that have the latent capacity to produce chondromucin. Chondroma, the benign form of these tumors, is characterized microscopically by a typical and unique structure. It consists of groups of two or more cartilage cells lying in a homogeoneous matrix of chondromucin. The tumor may be separated into lobular compartments by strands of fibrous connective tissue. The appearance of the rapidly growing or sarcomatous variety of cartilaginous tumor is more subject to variation than

is that of the benign form. In the former all gradations in the development of the type cell from the most immature phase to the fully adult cartilage cell may be seen in a single microscopic field. The undifferentiated cells are spindle-shaped, while those in the zone nearer the adult or fully differentiated cartilage cells are polymorphic. The blood vessels occur in the stroma of the connective tissue and are usually few and poorly formed.

Although, as we have mentioned previously, simple chondrogenic or osteogenic tumors occur rarely in chickens, connective tissue tumors, usually of the malignant type, occasionally occur in which cartilage or bone or both may be present. In these instances tumors that are primarily and predominantly fibrosarcomas in structure have within them small to large single or multiple regions of chondrogenic or osteogenic tissue. Tumors may be found composed of immature fibroblastic connective tissue, cartilage, and bone. Such specimens may be designated fibrochondroosteosarcoma. Tumors composed of immature chondrogenic and osteogenic tissues also have been noted in chickens. The multipotency of the primitive fibroblast accounts for these unusual neoplastic combinations.

**Histiocytic sarcoma.** This type of connective tissue tumor was first described adequately and established as a definite neoplastic entity by Jackson (1936a).[3] He reported having encountered fourteen cases among 203 cases of neoplasia of poultry. In our material histiocytic sarcoma has been observed sufficiently often to suggest that this is a frequently occurring connective tissue tumor of chickens.

In our series of eleven cases the ages of the birds varied from 35 days to sixteen months, with seven of them aged one year or less. While it would appear that histiocytic sarcoma may arise from almost any situation in the body, the ovary seems to be one of the sites of predilection. Other sites in which histiocytic sarcoma has occurred include esophagus, wattle, breast, pectoral muscle, subcutis, liver, and spleen. Campbell (1943) described a case primary in the kidney. Other organs, such as the lungs, gizzard, and heart, also have been involved, but involvement of these organs and of the serous tissues of the abdomen probably represents secondary rather than primary manifestations of the neoplasm.

The gross appearance of histiocytic sarcomas is not diagnostically characteristic. Encapsulation does not occur. These tumors are invasive and extend into the surrounding tissues in a rather diffuse manner. Those that arise in the ovary may be expected to extend to the contiguous tissues, especially the serosa, and to fuse the intestines, mesentery, and pancreas into a single unit. One should keep in mind, however, that similar spread by

---

[3] Those interested in the histogenesis of histiocytic sarcoma as exemplified by the so-called Rous sarcoma should consult McGowan (1928), whose views are, in general, in agreement with those of Jackson (1936a).

implantation is rather characteristic of other types of malignant ovarian neo-
plasia. Distant metastasis of histiocytic sarcoma to the lungs may occur, and
occasionally microscopic foci may be found in the kidneys. The metastatic
growths in the lung are often severe, and both lungs may appear as solid
masses of neoplastic tissue. All histiocytic sarcomas are potentially malignant
and may be expected to set up secondary foci if the primary process continues
long enough.
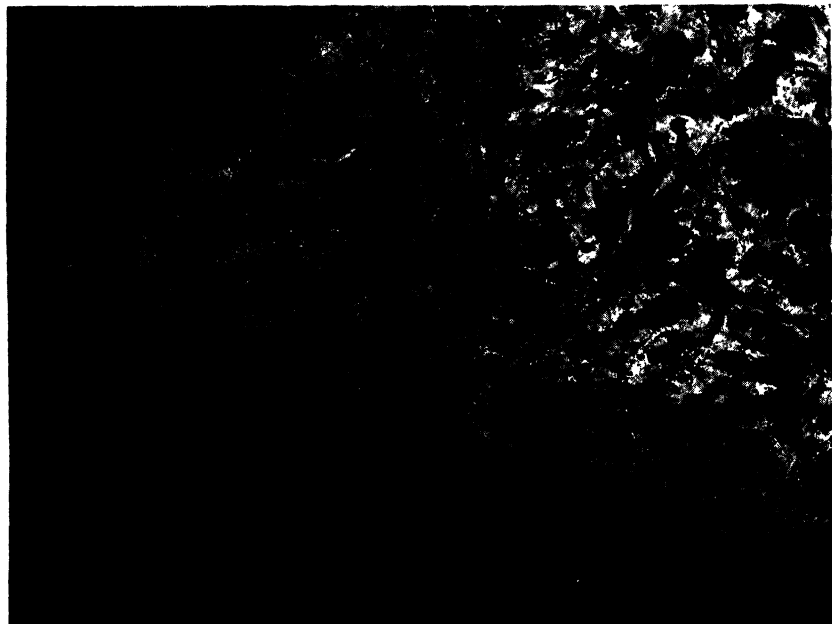


Fig. 30.3. Histiocytic sarcoma of the spleen. The varied character of the cellular con-
stituents is shown. ×285.

Microscopically, to the inexperienced, histiocytic sarcomas are likely to
present a structure of confusing complexity. These tumors usually appear
microscopically as a mixture of two or more types of cells that, while morpho-
logically dissimilar, are in fact closely related histogenetically (Fig. 30.3).
The cells that may be recognized are (1) spindle-shaped cells that usually
appear in groups or bundles somewhat like the structure of simple fibro-
sarcoma; (2) stellate reticulum-producing cells (fixed histiocytes) ; and (3) ·
large phagocytic cells or macrophages (free histiocytes), the latter usually
showing evidence of their functional specificity. In addition, one usually can
observe numerous transitional forms, of which many are polymorphic. ·
Foreign body giant cells are fairly often present in regions where macro-
phages are numerous. In those instances in which the tumor has metastasized,
there is usually present in the primary situation a greater number of spindle-
shaped cells than there are macrophages or histiocytic forms. In metastatic

foci the reverse is usually true, and macrophages and the primitive histiocytic forms predominate.

The microscopic diagnosis of histiocytic sarcoma is facilitated if the varied character of the type cell is kept in mind. The cellular constituents vary from spindle-shaped elements resembling fibroblasts to stellate and polygonal elements of bizarre form. A mixed type of structure is often the most striking feature of the microscopic picture. The recognition of the various constituents comprising these so-called mixed tumors is sufficient to distinguish their true character.

**Neurogenic sarcoma.** Neoplasia of connective tissue elements of peripheral nerve trunks has been reported by Jackson (1936a), Olson and Bullis (1942) and Duran-Reynals (1946a). Jackson observed such tumors as multiple nodules usually associated with the cutaneous nerves. Olson and Bullis described five cases. In two cases the tumor was single, and involved the ganglia of the dorsal root of the brachial nerve plexus (Fig. 30.4); in one case the tumor was found in two widely separated nerve trunks; and in the remaining two cases the lesions were not localized within the nerve but infiltrated the tissues adjacent to the affected nerves.
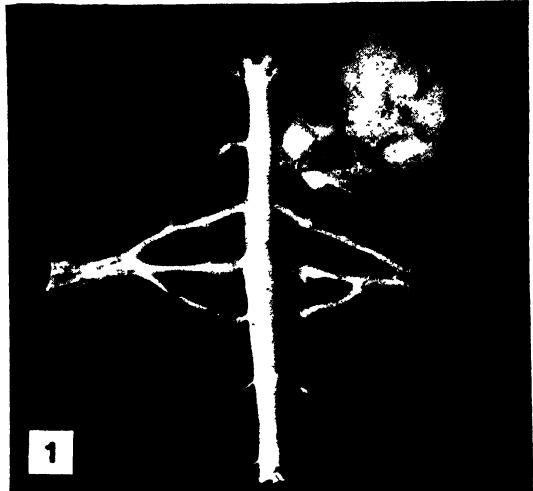


FIG. 30.4. Neurogenic sarcoma apparently arising from the dorsal root ganglion of the first thoracic spinal nerve.

The minute structure of the tumors was quite similar and consisted of fibroblastic elements of low malignancy which showed a distinct tendency to assume a whorl-like arrangement, sometimes with fissures. The histogenesis of these tumors is rather obscure, but it seems probable that they may originate from the fibrous sheath of the nerves.

Proper classification of these tumors is difficult. The whorl-like formations are similar to multiple neurofibromatosis of man, but except for Jackson's case the localization and invasiveness of these tumors are dissimilar from the characteristics of neurofibromatosis in human beings. A palisading of nuclei often noted in neurogenic sarcoma of human beings was not observed in any of the cases in chickens. Future study of a larger group of such cases may be expected to clarify the identity of these tumors and perhaps provide a more suitable classification.

*Metastasis and malignancy.* Since the type cell of the benign and malig-

nant forms of the connective tissue tumors is fundamentally the same, all of
these tumors are potentially malignant.  However, it is recognized that many
grow slowly, never invade the surrounding tissues, and continue indefinitely
as strictly localized processes.  It is our impression that in chickens the benign
forms of connective tissue neoplasms occur much less frequently than those
that are malignant.  The malignant forms are capable of widespread metasta-
sis with secondary foci occurring in the visceral organs.  The most striking
metastatic manifestations are those of the lungs and of the intestinal serosa.
In the histiocytic sarcoma there is some evidence that what may appear to be
secondary foci of metastatic origin are in fact independent tumors arising as
a result of a process that is systemic rather than local.

Diagnostic characteristics.  The features that aid in the diagnosis of the
connective tissue tumors are those associated with derivatives of the mesen-
chyme.  The recognition of intercellular fibrils, reticulum, collagen, mucin,
cartilage, or bone will suggest the connective tissue origin of these tumors.

### TUMORS OF MUSCLE TISSUE

Two general classes of tumors of muscle tissue are recognized.  Tumors of
one class, known as rhabdomyoblastomas, are composed of muscle cells that
have the inherent ability to produce within the cytoplasm both longitudinal
and cross striations.  Tumors of the other class, designated leiomyoblastomas,
are composed of smooth muscle cells and their associated elements.  Tumors
of either class may be either benign or malignant.  The benign form of
rhabdomyoblastoma is known as rhabdomyoma, the malignant form as
rhabdomyosarcoma.  Benign leiomyoblastomas are designated leiomyomas,
while those that are malignant are known as leiomyosarcomas.

A review of the literature indicates definitely that rhabdomyoblastoma is
among the rarer forms of neoplasms of chickens.  So far as we know, only a
few cases have been reported.  Meyer (Feldman, 1932) reported a rhabdo-
myoma that arose in the skeletal musculature of the sternum of a chicken.
The tumor was multiple and consisted of six separate masses measuring from
1 to 2 cm. in diameter.  They were composed largely of striated muscle fibers,
partially separated by strands of connective tissue.  On account of the mixed
character of the growth, Meyer designated the tumor "fibromyoma striocellu-
lare."  Another case of rhabdomyoblastoma was that reported by Peyron and
Blier (1927).  The tumor arose in the region of the hip joint of a rooster,
grew slowly, and proved transplantable.  Babic (1931) described rhabdo-
myomas of the pectoral muscle and submental region in a chicken.  Olson
and Bullis (1942) found two cases of neoplasia which were diagnosed
rhabdomyoma.  In one bird the tumors were multiple with a peculiar bi-
lateral symmetry of occurrence.  In the other case a small tumor was found
in the semitendinosus muscle with a secondary nodule on the intestine.  In

the absence of material for study, further description or discussion of this class of muscular tumors is omitted. In the remarks that follow, tumors of smooth muscle only are considered.

*Frequency of occurrence.* Tumors composed of the elements of smooth muscle are among the commoner neoplasms of chickens. In Jackson's (1936a, p. 432) series of forty-three primary tumors of the female reproductive system, fifteen were leiomyomas. Among 384 neoplasms of chickens encountered by Olson and Bullis (1942), thirty-four were leiomyomas.[4] Nelson (1946) found fifty-nine cases in a breeding flock of 1,108 hens all of which were subjected to necropsy examination.

*Sites of occurrence.* Although leiomyoblastomas may arise from smooth muscle wherever the tissue occurs, the vast majority of the tumors occur in the ligament of the oviduct or in the oviduct proper. Other situations in which tumors of smooth muscle have occurred include the muscular walls of the large and small intestines, the mesentery, the gizzard, and the crop.

An unusual case was described by Jackson (1936a, p. 332) in which a mass weighing approximately 1 kg. and designated leiomyoma fibrosum involved the ovary. Multiple tumors of similar character affected also the ovarian bursa and the proctodeum. In another case mentioned by Jackson, a leiomyoma involved the ovary and the oviductal ligament.

*Effects on the host.* Most tumors composed of smooth muscle apparently grow slowly and require considerable time to attain sufficient size to interfere seriously with the normal functioning of the involved or adjacent tissues. Since these tumors are seldom invasive or destructive locally, the ultimate effect, if any, that they may exert on the host will depend largely on the amount of mechanical interference their presence may have on proper functioning of the involved parts. Those of the oviduct and of the broad ligament could conceivably reduce or preclude egg production. Actually, Olson and Bullis found that eighteen birds that had leiomyoma of the oviduct or mesosalpinx were more than average in egg-laying ability. The interval between the last egg laid and necropsy varied from 1 to 73 days and averaged only 12 days. A causal relation between a long period of heavy egg production and development of leiomyoma of the mesosalpinx was suspected by Olson and Bullis. Nelson's (1946) data do not indicate a strong familial tendency for the disease. Leiomyomas of the intestines or other hollow organs might provide obstruction to the free passage of ingesta. If the tumor is malignant and secondary subserous implantations have occurred, ascites may develop.

While the foregoing possible effects may ensue as a consequence of the presence of tumors of smooth muscle, it should be recognized that, owing to

---

[4] The 384 tumors listed by Olson and Bullis included 213 lymphocytomas. Cases of leukosis were included.

the slow rate of growth, these tumors may be present for prolonged periods without giving rise to objective symptoms.

*Gross and microscopic description.* Most leiomyoblastomas are smooth, elongated, ovoid, or irregularly spherical tumors. The benign forms usually are covered with a capsular structure that can be removed with difficulty. These tumors are resilient and of firm consistency. The size varies from 1 cm. or less to large masses several centimeters in diameter. They are flesh pink to grayish white, and when they are cut across, the structure frequently appears distinctly fibrous. Although some specimens have a pedunculated form of attachment to the tissues from which they arise, in most instances these tumors are attached firmly by a rather broad base or over a considerable portion of their structures to the adjacent tissues (Fig. 30.5).

Microscopically, a typical leiomyoma presents a compactly knit structure composed of neoplastic smooth muscle cells. The degree of cellularity varies somewhat with the rate of growth, the
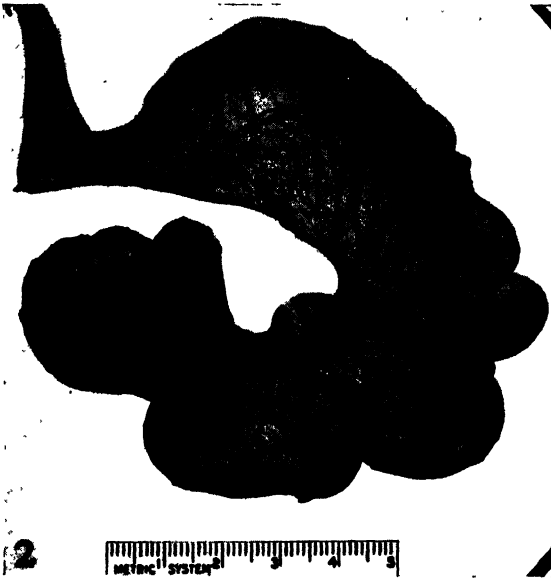


FIG. 30.5. A relatively small leiomyoma in the mesosalpinx.

cells being most numerous in those tumors in which the rate of growth is accelerated. Groups of cells usually are arranged in units of bundles which are disposed in every conceivable direction. The nuclei are ovoid or often elongated and contain a considerable amount of granular chromatin. The amount of connective tissue present between the respective bundles of muscle cells is subject to considerable variation. In some regions it is unrecognizable, while infrequently the fibrous elements may equal in amount the muscular tissue. Vascular channels are usually numerous.

In leiomyosarcoma the cells have the appearance of immaturity and are more numerous than in the slowly growing benign form. Cells undergoing mitosis are seen frequently, and the invasive tendencies of the process usually can be distinguished (Fig. 30.6).

*Metastasis.* As mentioned previously, tumors composed of smooth muscle cells are seldom malignant, and consequently metastasis is observed infre-

quently. Even those that appear malignant morphologically are slow to metastasize. Should metastasis take place, the liver and the lungs are the most likely sites of secondary foci. There is some evidence that occasionally smooth muscle tumors occur that arise multicentrically. In these, the absence of mitosis or other signs of immaturity of the cells indicates that the multiple tumors represent multiple expressions of a similar process rather than metastasis.
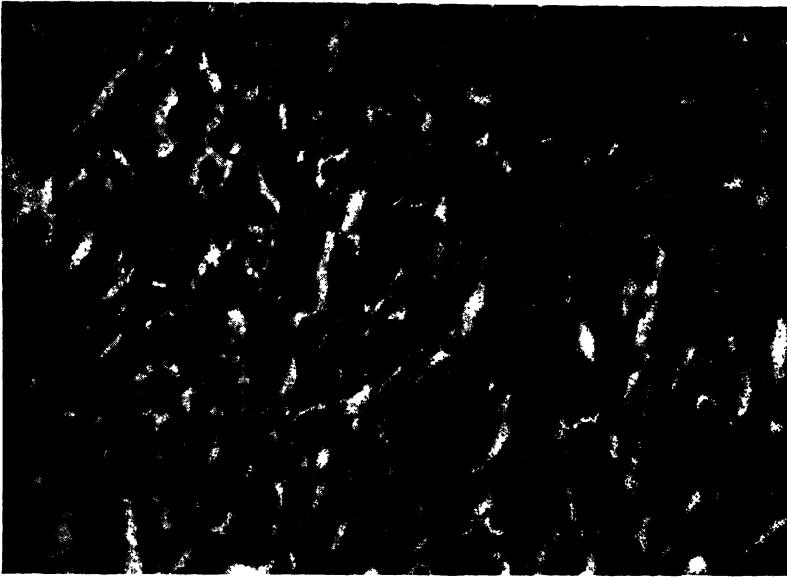


FIG. 30.6. Malignant leiomyoblastoma (leiomyosarcoma) of the oviduct of a 2-year-old hen. ×660.

*Diagnostic characteristics.* Tumors that arise in intimate association with tissues or organs containing smooth muscle are likely to be leiomyoblastomas. This is especially true of tumors of the wall of the oviduct, the ventral ligament, or the walls of the intestines. The presence of smooth muscle cells arranged in interlacing strands or bundles disposed in a divergent manner is fairly characteristic. In differentiating tumors of smooth muscle from certain connective tissue tumors, the van Gieson stain may be helpful. The presence of myoglia is specific for identifying smooth muscle cells. However, to stain tissue for myoglia requires fixation in Zenker's solution and staining by phosphotungstic acid hematoxylin.

## TUMORS OF BLOOD AND LYMPH CHANNELS

*General considerations.* Neoplasia may arise from the elements of the blood and the lymph vessels. Tumors developing from neoplastic growth of

blood vascular channels are called hemangiomas and those from lymph channels lymphangiomas. Different types of growth may occur. In some instances the newly formed vessels are small and capillary-like, and the terms "capillary hemangioma" or "capillary lymphangioma" are applicable. In other tumors the blood or lymph spaces are large, and cavernous hemangioma or cavernous lymphangioma is the appropriate term. The tumors may be benign or malignant. In some hemangioblastomas, particularly those which are malignant, parts of the cellular mass may not show any evidence of differentiation and may appear similar to a fibroblastic sarcoma. The general character of such tumors is revealed, however, by the more differentiated regions in which distinct vascular channels are formed. Such tumors require careful study for correct identification.

Jackson (1936a, pp. 17–18) pointed out the difficulties in connection with the term endothelioma, which sometimes is used in connection with tumors of vascular channels. The lumen of both normal and neoplastic vascular channels is lined with specialized cells called endothelial cells. Whether these are derived from the same source as the angioblast forming the vessel wall or have an independent origin is a moot question. While some tumors have been designated simply as endotheliomas, their exact character, except in certain cases, is extremely vague. Such a diagnosis might refer to a tumor of the lining cells of blood channels. It might refer to a tumor of the meninges of the brain or spinal cord. It might also refer to a tumor arising from the reticulo-endothelium which is disseminated so widely in all organs of the body that the existence of such a specialized tissue is not often recognized. In actual practice, the term endothelioma usually can be avoided by careful study of the material which enables one to arrive at a proper classification. Thus, meningeal tumors of this type are properly a form of fibroblastic neoplasia. Those of the reticulo-endothelial system are more properly associated with neoplasia of the hemopoietic organs. A tumor of the endothelium of vessels would be extremely difficult to recognize unless there was sufficient differentiation to form capillaries, in which case the origin of the cells would be in question and, furthermore, the tumor probably would be recognized as a capillary hemangioma.

**Blood vascular tumors.** *Frequency of occurrence.* Tumors of blood vascular tissue appear to be relatively infrequent in the chicken as judged by reports in the literature. Heim (1931) found five cases in the literature he reviewed. These included four cases of multiple hemangioma reported by Schurman and Pauly. The tumors affected the skin, musculature, mesentery, serosa of the duodenum, kidney, lungs, and subcutis of the throat. Babic (1931) described four cases of hemangioma in chickens. These involved the liver in two birds and the skin and subcutis of the head and the eyelid in another chicken. In the fourth case the tumor was a cavernous hemangioma

on the peritoneum. Babic also described an angiomatous nevus of the wing in a canary. Olson and Bullis (1942) found five instances of hemangioma in a collection of 384 tumors of chickens. Three were of the cavernous type, and two were capillary hemangiomas. Only two of the tumors were considered malignant. The liver was affected in four of the five cases, suggesting a predilection of the liver of the chicken for the development of hemangiomas. In two cases, the tumor in the liver was so small that it easily might have been overlooked. Ball (1945) reported a hemangio-endothelioma of the iris in a 24-week-old pullet.

*Gross and microscopic description.* Hemangioblastomas are variable in appearance according to the type of tumor. The large cavernous form is characterized by greatly distended blood spaces whose lining is a thin wall of endothelial cells (Fig. 30.7). The distended blood spaces often protrude



FIG. 30.7. Cavernous hemangio-endothelioma of the mesentery. ×70.

from the affected organ or tissue. Such tumors are typical in macroscopic appearance. Capillary hemangiomas may appear as solid masses of neoplastic tissue varying from gray-pink to red. Histologically, their character is apparent. Small capillaries containing blood are the essential features. All gradations between the cavernous and capillary forms may occur.

**Lymph vascular tumors.** Tumors of the lymph vascular elements may possess the same histologic features as those of the blood vascular system, except that lymph instead of blood is contained within the spaces formed by the tumor cells. A few blood cells may be found occasionally in the lymph.

Macroscopically, milky fluid (lymph) may be recognized in the cystlike structures of cavernous lymphangioma.

Very few cases of lymphangioma have been described in the chicken. Heim (1931) mentioned one case of lymphangioma of the eyelid in a bird which was found by Teutschlaender. Babic (1931) described a lymphangiolipoma of the mesenteric serosa in a chicken. Olson and Bullis (1942) encountered one case of lymphangioma in which the tumor was a pedunculated mass attached to the ovary.

**Telangiectasis.** Telangiectasis may be considered as a benign form of hemangioblastoma in which a group of blood or lymph vessels become dilated with blood or lymph. The pathogenesis of telangiectasis is not known, and different authors have suggested that it is either a congenital or a hereditary disease or that it follows injury and repair to an organ in which the circulation has been disturbed (Ewing, 1928; Feldman, 1932). Telangiectasis may be multiple and capillary or cavernous.

### TUMORS OF HEMOBLASTIC ORIGIN

**Lymphocytoma.** *Definition and terminology.* The neoplastic lymphocyte is the type cell of a common tumor of chickens to which the simple descriptive term of lymphocytoma may be applied. Many other terms have been given to this tumor. These include round-cell sarcoma, lymphadenoma, lymphocytomatosis, lymphomatosis, lymphatic leukosis, leukoblastic leukosis, leukemia, and others. The question of terminology is made somewhat difficult because of the frequent association of lymphocytoma with fowl paralysis and fowl leukosis. This has come about as a result of experiments intended to demonstrate the transmissibility of these diseases. At the present time fowl leukosis is the only one of these entities in which transmissibility has been established definitely. The experimental transmission of lymphocytoma and fowl paralysis is an unsettled question since research has yielded conflicting results. A recent report by Davis and Doyle (1947a) provides a concise review of the literature on this question and shows the diverse conclusions reached from attempts at experimental transmission experiments. Work in the future may be expected to decide this point, and for the present, etiologic considerations should not be allowed any part in the nomenclature of these diseases. We have adopted the precept of dealing only with the pathologic anatomy in assigning names to these conditions. Further work may indicate that what we today group under the term lymphocytoma is a composite of several entities. Since the study of lymphocytoma of the chicken is in its infancy, we may anticipate a modification of certain of our views concerning this condition.

*Histogenesis.* The lymphocyte is a most important cell in the animal organism and has many diverse physiologic functions. Our knowledge of

these functions is far from complete. The lymphocyte is an integral part of the widespread reticulo-endothelial system. It is normally an unstable cell and may assume many different forms. Jordan (1936) expressed the belief that the lymphocyte is a hemoblast capable of developing into any type of blood cell. According to him the large lymphocyte as found in the circulating blood represents a young cell with the foregoing potentialities. The small lymphocyte is a more mature adult cell which has lost or outgrown the ability to become transformed into other types. The factor or factors which govern and direct the transformation of lymphocytes into other cells are not well understood.

The histogenesis of lymphocytoma is likewise obscure. The orthodox concept of neoplasia that a tumor begins as a result of a cell or a localized focus of cells assuming a state of neoplasia does not seem applicable in the case of lymphocytoma. Lymphocytoma appears to be more of a systemic disease in which the lymphocytes in widely scattered regions become transformed simultaneously into neoplastic cells. This feature of lymphocytoma suggests the existence of a specific stimulus which, when applied to lymphocytes in a susceptible stage, causes them to become capable of the unrestricted growth characteristic of autonomous proliferation. However, such a stimulating agent has yet to be demonstrated. Lymphocytoma is found more often in certain organs rich in lymphoid tissue than in others. Although it commonly affects the liver, gonad, kidney, and spleen, other organs likewise rich in lymphoid tissue, such as the marrow, bursa of Fabricius, and thymus, are affected less commonly. This fact suggests that the lymphoid tissue in the latter group of organs is in a functional state different from that in the organs first mentioned, rendering it less susceptible to the hypothetical stimulus for neoplasia.

Structurally, three forms of lymphocytoma may be recognized. These are the diffuse, the nodular, and the combined diffuse and nodular forms. In the diffuse form the affected tissues are infiltrated diffusely with neoplastic lymphocytes which crowd and replace the parenchymatous tissues of the involved organs. The nodular form is characterized by a follicular arrangement of the tumor cells, which are confined by a more or less well-developed retaining wall of connective tissue, rich in reticulum. The third form of lymphocytoma is a combination of the diffuse and the nodular form. In such cases a single organ may show both forms, or one organ may be affected with one form and other organs with the other form.

A possible explanation has been suggested for the existence of these diverse forms of lymphocytoma (Olson and Bullis, 1942) based on the hypothesis that they develop because of the inherent resistance on the part of the individual host to the growth of the tumor. Thus diffuse lymphocytoma develops in birds that have little resistance to the growth of the

tumor, and nodular lymphocytoma develops in birds which are able to muster considerable resistance. The third form of combined diffuse and nodular lymphocytoma develops in birds that have only a moderate degree of resistance to growth of the tumor. Evidence in support of this hypothesis is provided by several facts. Involvement of fewer organs and less extensive damage usually are noted in the nodular form of disease than in the diffuse form. Birds affected with the nodular form are often emaciated, suggesting a prolonged course, whereas the carcass in cases of the diffuse form is usually well nourished. The response of connective tissue in the nodular form suggests an attempt to limit the growth of the tumor.

Solution of the problem of the causation of lymphocytoma is an almost essential requirement for developing a sound conception of the histogenesis of the disease. For the present it may be said that lymphocytoma develops from lymphoid cells which are scattered widely throughout the body, but the stimulus or mechanism by which the cells become malignant is yet unknown.

*Frequency.* Lymphocytoma is without question the most common form of neoplasia affecting the domestic chicken. As discussed in the section dealing with general incidence of neoplasia, precise data on the frequency of occurrence of tumors are difficult to obtain. Olson and Bullis (1942) found 213 cases of lymphocytoma in a collection of 384 tumors, an incidence of 55.5 per cent. It has been recognized by poultry pathologists that lymphocytomas may be much more common in some flocks than in others. They may be so common as to constitute a serious economic problem in some flocks. Experimental results such as were reported by Hutt, Cole, and Bruckner (1941) suggest that the incidence of neoplasia can be controlled by selective breeding. Since they stated that "approximately 95 per cent of the deaths attributed to neoplasms were caused by lymphomatosis of one kind or another," it is inferred that by selective breeding lymphocytoma would be affected significantly. The exact effect of such a breeding program on the incidence of lymphocytoma alone is unfortunately not available, since suitable data on which to draw conclusions are not given.

The disease usually affects chickens less than a year old. Birds that had lymphocytoma in the series of cases studied by Olson and Bullis had an average age of about eight and a half months. The youngest bird affected was six weeks old and the oldest was two years of age. It was formerly believed that males were affected about as often as females when one considered the disproportionate number of male and female chickens in the general poultry population. The data of Olson and Bullis tend to suggest that the male is affected with lymphocytoma less commonly than is the female. Burmester (1945) reported that the incidence of lymphomatosis (presumed to include lymphocytoma, neurolymphomatosis, and the iritis commonly associated with neurolymphomatosis) was twice as great in female as compared to male

uninoculated chickens. Marine and Rosen (1940, 1941) noted a rather high incidence of lymphomatosis (which in most of their cases appears to be similar to lymphocytoma) in castrated male chickens. They suggested that an imbalance of hormones may have activated a latent tumor-producing agent in these birds. A higher incidence of lymphomatosis in castrated male chickens was also observed by Burmester and Nelson (1945) whose studies showed a similar effect in castrated females. They further reported that administration of a female sex hormone (diethylstilbestrol) lowered the incidence in capons but had no effect on male chickens. Administration of a male sex hormone (testosterone proprionate) seemed to lower the incidence of lymphomatosis in both males and females. Burmester and Nelson (1945) suggest that these hormones increase the resistance of the bird to lymphomatosis. Davis and Doyle (1947a) also reported a higher incidence of visceral lymphomatosis in females and capons than in male chickens.

*Anatomic situation.* Lymphocytoma may affect nearly every organ or tissue in the chicken, and in a given case usually more than one organ or tissue is affected.

In a series of 213 cases of lymphocytoma (Olson and Bullis, 1942) there were only thirty-one instances in which the disease was confined to a single organ or tissue. The liver, spleen, kidney, and gonad were the organs most commonly found to be affected. Nineteen different organs or tissues, exclusive of nerves and the circulating blood, were found to be involved with the disease. In this series the different combinations of organs or tissues that might be affected with lymphocytoma were studied. The great variation of the manner in which lymphocytoma may express itself is indicated by the fact that 152 different combinations were found. The more frequent of these combinations were as follows: ovary alone (ten cases) ; liver, spleen, kidney, gonad, and marrow (seven cases) ; liver, spleen, and kidney (seven cases) ; liver and spleen (five cases) . The ovary only was affected in ten cases and the peritoneum only in five cases. The histologic form of the disease appears to be a determining factor with respect to the number of organs or tissues involved. The diffuse and the combined nodular and diffuse forms tend to be more widespread and affect more organs than does the nodular form.

In view of the tendency of lymphocytoma to become widely disseminated, it is logical to assume that nerve tissue may be affected with the disease. In eighty-three of the 213 cases of lymphocytoma studied by Olson and Bullis (1942) , deposits of neoplastic lymphoid tissue were found in the peripheral nerves. The amount of tumor tissue varied from a localized, lightly infiltrated region to complete replacement of the nerve tissue and marked enlargement of the affected nerve. The presence of such deposits of lymphoid tissue associated with peripheral nerves raises the question whether they represent foci of lymphocytoma or are an expression of the disease known as

fowl paralysis. The lesions of the nerves in fowl paralysis have been described as either inflammatory or neoplastic. When inflammatory, the lesions consist of polyblastic infiltration (lymphocytes, histiocytes, and plasma cells) often associated with the proliferation of the cells of the sheath of Schwann and degeneration of neurons in the ganglia. The neoplastic lesions differ from the inflammatory lesions in that the infiltrating lymphoid cells have a definitely neoplastic character, are multiplying actively, and may be so aggressive as to replace almost entirely the nerve elements within the sheath. The neoplastic process may penetrate the nerve sheath and infiltrate the adjacent surrounding tissue. Separation of the lesions into two such groups is complicated by those cases in which both types of lesions are found.

Involvement of the nerves in some cases of lymphocytoma is obviously due to the penetration of the nerve sheath from without by neoplastic lymphoid tissue. However, those instances of lymphocytoma in which neoplastic lymphoid tissue is found in nerves at a site removed from other tissues affected with lymphocytoma constitute a difficult problem in interpretation. They may represent metastasis of the tumor from a primary focus situated elsewhere. The tumors may have developed in the nerve in response to a hypothetical causative agent of lymphocytoma. They may be lesions of fowl paralysis existing coincidentally with lymphocytoma. On the other hand, one may assume that a single agent is responsible for both fowl paralysis and lymphocytoma and that the type of response to the agent depends on factors as yet unknown. A final solution of this question must await the solution of the problem of causation.

*Effects on the host.* Lymphocytoma usually is considered as a fatal disease. While this may be generally true, in rare instances a bird may recover from the disease. In our material two such cases have been noted in which the birds had multiple tumors of the skin. Diagnosis of the disease was made by biopsy and histologic examination of representative cutaneous lesions. The birds were held under observation for several weeks, and the remaining tumors of the skin disappeared. A similar regression of a tumor in a visceral organ might occur, but the recognition of such a case would be extremely difficult. Recently, Davis and Doyle (1947b) have reported a study of monthly liver biopsies on ninety-six chickens done over a ten-month period. Some of the birds had been inoculated with material from a case of spontaneous visceral lymphomatosis and others were uninoculated controls. These interesting data showed that fatal cases of the disease developed very rapidly. For example, biopsy material was normal in birds that died with lymphomatosis three to four weeks after biopsy. In some instances, lesions of the liver characteristic of lymphomatosis were observed in biopsy material and the lesions later disappeared, indicating recovery from the disease.

There are no specific or pathognomonic symptoms displayed by birds affected with lymphocytoma. In many instances birds under relatively close

obervation may die with the disease without indication of ill health. In most cases signs of a general disturbance of physical health are evident for a variable period preceding death. These signs are listlessness, inappetence, ruffling of the feathers, and general depression. An affected bird is often first noted to be standing in droopy attitude with its eyes closed and with an intermittent shaking of the head as though the sensorium were befogged. The location of the lesions may provoke distinctive signs referable to their situation. For example, tumors of the skin or musculature cause localized swelling, involvement of the digestive tract may cause either diarrhea or obstipation, and involvement of a nerve may cause paralysis of the part supplied by the nerve. Palpation of the abdomen may reveal displacement of the viscera due to an enlarged liver. Emaciation will develop in cases of long standing, which are usually due to the nodular form of lymphocytoma.

Olson and Bullis (1942) obtained data on the egg production of fifteen birds that died of lymphocytoma. These birds were considered average to slightly better than average producers of eggs. The rather rapid course of lymphocytoma is suggested by the finding of a relatively short interval between cessation of egg production and the death of the birds. This period averaged 38 days and varied from 4 to 73 days.

Olson and Dukes (1938) found that the basal metabolic rates of two chickens affected with lymphocytoma were greatly increased over the normal level. In this respect these cases of lymphocytoma of the chicken were similar to neoplastic diseases of the lymphoid cell system as encountered in human beings. The rate of basal metabolism should be studied on more cases of the disease, and if it is found that an increase is a constant feature, the result might possibly explain the rapid wasting and emaciation asociated with the more chronic form of lymphocytoma.

*Gross and microscopic description.* Organs or tissues affected with lymphocytoma have a gross appearance which varies with the extent of infiltration and the character of the process. Organs which on gross examination appear normal may contain neoplastic foci when examined microscopically. The characteristic color of neoplastic lymphoid tissue is gray-white, and the tissue may have a red tint in the more highly vascular areas. Necrosis of the tumor substance is not observed commonly but may develop in regions of the tumor in which the blood supply has been reduced by occlusion of the vessels either from pressure or from the infiltrative growth of the neoplasm.

As mentioned previously, the growth may be diffuse or nodular or a combination of the two. In diffuse lymphocytoma the affected organs are enlarged uniformly, and the color of the organs may change until it resembles the gray-white of the tumor. The extent of enlargement and the degree of change of color depend on the amount of tumor tissue present. Organs severely affected and in which there is much replacement of the parenchyma are quite soft. In nodular lymphocytoma the neoplastic tissue has a well-

developed supporting framework of connective tissue which adds much to the firmness of the tumor. Affected organs contain nodular gray-white masses whose margins are discrete and sharply defined. The nodules may almost completely replace the parenchyma of an organ, reducing the latter to narrow bands compressed between the masses of tumor (Fig. 30.8). In some cases of nodular lymphocytoma, the tumor may be distributed throughout the affected organ and resemble the diffuse form of the disease. Such cases can be recognized by the firmer consistency of the tumor and its histologic ap-
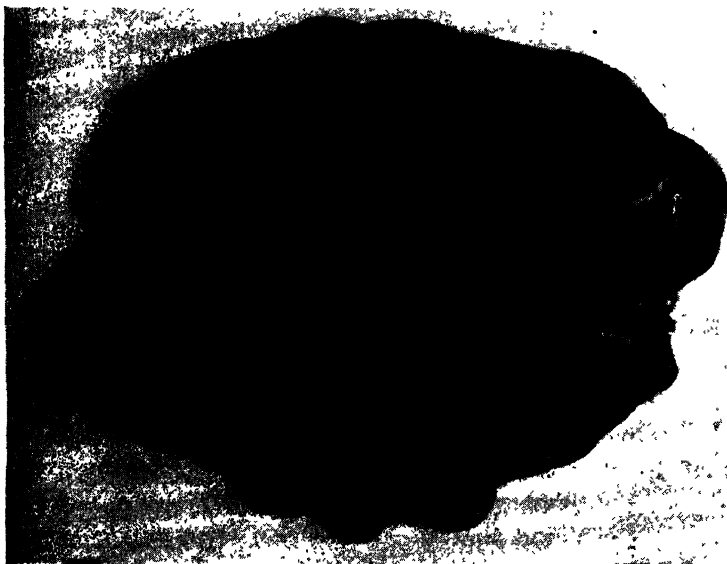


FIG. 30.8. Lymphocytoma; nodular type, showing multiple lesions in the liver.

pearance. Fairly frequently the tumor may erode the walls of the blood vessels and cause hemorrhage. Fatal hemorrhage into the peritoneal cavity may occur from rupture of the taut capsule of organs greatly enlarged from growth of the neoplasms. Such hemorrhages are noted most often from the liver and spleen.

Microscopically, the tumor consists of masses of proliferating neoplastic lymphoid cells situated extravascularly (Fig. 30.9). The foci of cells tend to develop most rapidly in the immediate vicinity of blood vessels. The cells of the tumor are quite uniform and comparable in size to the large lymphocyte or monocyte of the circulating blood. They tend to be spherical, although in the denser parts of the tumor they are so compact that the shape either cannot be distinguished or is distorted. The cytoplasm is relatively scant, is without specific granulation, and stains faintly blue with hematoxylin and eosin. The nucleus is relatively large and has a vesicular appearance. The chromatin is arranged as an irregular band at the nuclear margin

and in small clumps in the nucleoplasm. One or two distinct nucleoli are usually present. Mitotic figures are commonly found. A fine meshwork of reticulum enclosing small groups of tumor cells can be demonstrated by appropriate histologic procedures.

In the diffuse form of lymphocytoma, the infiltration of the tumor between the parenchymatous cells of an affected organ appears to proceed without the slightest restraint. As the disease progresses, the tumor cells destroy and replace the normal cells of the organ. This process may continue until the affected organ is almost completely converted into a solid mass of tumorous tissue.



FIG. 30.9. Lymphocytoma of the ovary. Ovarian tissue has been entirely replaced by the neoplasm, which consists of small irregular nests of lymphoid cells. ×45.

In nodular lymphocytoma a marked response of connective tissue accompanies the proliferation of neoplastic lymphoid cells. The connective tissue surrounds and isolates clumps of tumor cells, forming a sort of capsular wall. These foci of tumor cells may be small and isolated, but more often they are contiguous to other such foci. Sometimes several such foci seem to merge with one another, forming larger masses surrounded by a thicker wall of connective tissue. The fibroblastic components of the connective tissue wall are not anaplastic and would seem to be the response of the host attempting to delimit the growth of the tumor rather than a part of the tumor itself. Cases of lymphocytoma occur in which both the nodular and the diffuse form of the disease may be found in either the same organ or different organs of the same animal.

Occasionally, the cells of lymphocytoma may either erode or infiltrate into the lumina of blood vessels and thereby enter the circulation in numbers sufficient to be mistaken for a leukemic state, but this process is distinctly different from that leading to true leukemia. It should be regarded rather as an embolism.

In addition to such emboli of tumor cells, changes of the blood picture may be found occasionally in lymphocytoma. These are of a secondary nature. Foci of lymphocytoma are found fairly frequently in the bone marrow where they may disrupt normal hemopoiesis by mechanical means. For example, they may replace sufficient myeloid tissue to lead to a state of insufficiency, causing anemia and leukopenia. They may also mechanically dislodge unripe cells from the marrow, forcing them into the circulation.

The cellular changes of the circulating blood in birds suffering from
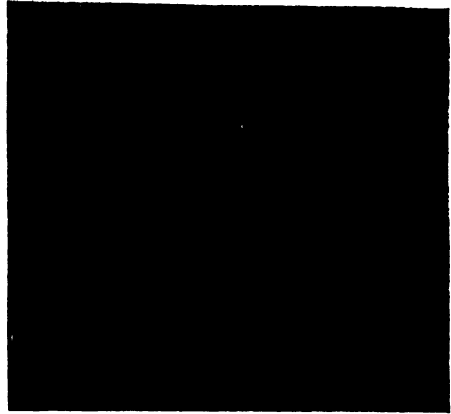
lymphocytoma have not received adequate attention. Part of this neglect is due to the difficulty of detecting cases sufficiently early so that the changes might be studied during the development of the disease. Several authors have made blood smear examinations of such cases at varying intervals preceding death, but their main objective was a search for pathologic cells in the blood rather than a study of variations of the cells normally present.

A fairly complete study was made of the blood in one of our cases, a lymphoid tumor which later was demonstrated to be transplantable (Olson, 1941). Although in this case the tumor has not been designated as lymphocytoma because of its transplantable nature, other features are such as would cause it to be considered as a lymphocytoma. These changes in the blood were observed during the 25-day period preceding death of the bird and may be summarized briefly as follows: Erythrocytes, hemoglobin, thrombocytes, eosinophils, and basophils were only slightly affected. The number of heterophils, lymphocytes, and monocytes fluctuated widely and approached normal levels only near the terminal stage of the disease. The severity of the involvement of the bone marrow did not seem sufficient to explain the variations observed. It seems probable that the disease in this instance was associated with the production of noxious materials which were responsible for the increase of the numbers of heterophils, lymphocytes, and monocytes.

*Extension.* The aggressive nature of lymphocytoma is revealed in many cases by the extensive lesions in markedly enlarged organs. As stated previously in the section on histogenesis, lymphocytoma is probably a systemic disease in which the process is initiated in several sites at the same time. Extension of the disease by direct spread from one tissue to another is illustrated by those cases in which the peritoneum is involved. The peritoneum may become affected by extension of the disease from the ovary, and the intestine may be involved in turn from the previously affected peritoneum. Further examples of such extension of the disease require only examination and study of material that comes to necropsy. Although emboli of tumor cells may be demonstrable in the blood vessels, acceptable evidence of true metastasis is difficult to obtain.

In this connection it is pertinent to mention a few unsuccessful attempts at autoplastic transplantation of lymphocytoma. In these experiments (Olson and Dukes, 1938) bits of skin tumor were transplanted into the subcutis and musculature of the birds from which the tissue was obtained for biopsy. Although only a few trials were made, in each case the implants failed to develop despite continued growth of the original skin tumors. Failure of such autotransplants may be due to concomitant immunity such as displayed by a transplantable lymphoid tumor in which the immunity developed 10 to 15 days after the initial graft protected the bird against subsequent grafts even though the original graft continued to grow (Olson,

1945). These experiments bear on the question of metastasis for they suggest that metastasis may be of rare occurrence in lymphocytoma. This would lead to the conception that extension of the disease is largely a matter of direct spread from one organ or tissue to another rather than of circulatory metastasis.

*Diagnostic characteristics.* With an adequate knowledge of the fundamental pathologic changes, it is usually a relatively simple task to differentiate lymphocytoma from other diseases. Myelocytoma, leukosis, fibrosarcoma, epithelioblastoma, and some types of granulomatous processes are some diseases which should be considered in arriving at a differential diagnosis of lymphocytoma. General features which may serve as a guide in distinguishing between these diseases are set forth in Table 2. Other less common tumors may cause confusion in the differential diagnosis of lymphocytoma. A good example is histiocytic sarcoma.

*Special features.* A neophyte delving into the literature pertaining to lymphocytoma will find a mass of conflicting information which serves to confuse rather than to clarify the situation. In the older literature, one will encounter the term "round cell sarcoma" with surprising regularity. While such a term is descriptive of the shape of cells found in such cases, it does not give any information on the histogenesis of the tumor. Heim (1931) made a thorough review of the literature on neoplasia of the chicken and, in addition to discussing "round cell sarcoma" of the connective tissue, devoted another section of his report to "round cell tumors of unknown genesis." This latter group was subdivided further into a "large celled" form and a "small celled" form. No doubt examples of what we today call lymphocytoma were included in both categories, as well as histiocytic sarcoma and perhaps other types of tumor.

The relation between spontaneous lymphocytoma and experimentally transmissible neoplasms of lymphoid cells is not well understood. Furth (1935) expressed the belief that the disease produced by the "Strain 2" tumor-producing agent studied by him is rare as a spontaneous disease of chickens and dissimilar from the commonly occurring lymphocytoma. Both Pentimalli (1941) and Olson (1941) have found a spontaneous lymphoid tumor that was transplantable to experimental chickens. The original cases of both possessed features which would permit them to be classified as lymphocytoma, and the principal reason for not doing so is that transplantability has not been demonstrated as a characteristic feature of lymphocytoma (Olson, 1940, 1942, 1947; Engelbreth-Holm, 1942; Duran-Reynals, 1946b). The factor or factors responsible for this discrepancy may become known some day and explain the situation. In the meantime this difference must be borne in mind. Some authors will differ with such a distinction and hold that lymphocytoma (lymphomatosis) is a transmissible disease etio-

TABLE 2

COMPARISON OF LYMPHOCYTOMA WITH OTHER DISEASES

| | Lymphocytoma | Myelocytoma | Leukosis | Fibrosarcoma | Epithelioblastoma | Granuloma |
|---|---|---|---|---|---|---|
| Age (average) | 9 months | 9 months | 11 months | 10 months | 12 months or more | Any age |
| Frequency | Common | Uncommon | Uncommon | Uncommon | Rare | Common |
| Course | Acute | Acute | Protracted | Variable | Variable | Variable |
| Location | Liver, gonad, kidney, and other tissues | Periosteum, liver, spleen, gonad, marrow, and other tissues | Bone marrow, liver, spleen, kidneys, recent hemorrhages may be present in fascia and intestinal mucosa | Any organ | Any epithelial tissue | Any tissue |
| Extent | Widespread or limited | Widespread or limited | Limited | Limited | Limited | Limited |
| Character | Nodular or diffuse | Diffuse | Diffuse | Localized | Localized | Localized |
| Texture | Soft or firm | Soft | Soft | Quite firm | Quite firm | Soft or firm |
| Color | Gray-white | Dull white | Organs pale | Gray-yellow | Not consistent | Yellow (necrosis common) |
| Blood | Usually normal, may show anemia, sometimes embolism of tumor cells | Anemia, myelocytes in circulation | Severe anemia, immature blood cells | Essentially normal | Normal | Sometimes leukocytosis |
| Histologic characteristics | Extravascular infiltrations with neoplastic lymphocytes | Extravascular infiltration of neoplastic myelocytes, cells may appear in blood stream | Intravascular collections of neoplastic unripe blood cells | Infiltration with neoplastic fibroblasts | Masses of neoplastic cells of epithelial origin | Inflammatory reaction |
| Causative factors | Unknown | Unknown | Filtrable agent | Unknown and filtrable agents | Unknown | Trauma, pathogenic bacteria, fungi, parasites, degenerative processes |

logically associated with fowl paralysis and leukosis. Such an association cannot be regarded as conclusively settled either one way or another except in the case of fowl leukosis, which the bulk of evidence indicates to be etiologically distinct from lymphocytoma and fowl paralysis.

Burmester and Prickett (1945) described strains of transplantable lymphoid tumors similar and apparently immunologically related (Burmester and Belding, 1947) to the lymphoid tumor reported by Olson (1941). Brewer and Brownstein (1946) state that "lymphomatous" liver and spleen material from several birds produced visceral lymphomatosis in young chicks. Details are given on only one strain which was infective with fresh affected tissues by feeding and simultaneous instillation in the eye and nose as well as subcutaneous inoculation. This tumor does not appear to be characterized by a definite rate of growth. Davis and Doyle (1947a) describe transmission of visceral lymphomatosis with a slower rate of growth than that observed with the lymphoid tumor by Olson. These transplantable lymphoid tumors and cases of visceral lymphomatosis had no predilection for growth in nerve tissue.

The association of spontaneous lymphocytoma with fowl paralysis is a feature still deserving of special attention. Some comments concerning this question have been made in this section, and other comments may be found in the chapter entitled "The avian leukosis complex."

**Myelocytoma.** The term "myelocytoma" was first applied to this disease by Pentimalli (1915) who described two cases. Ellermann (1920) recognized the condition in the course of his work with leukosis and called the disease aleukemic myelosis. Mathews (1929a) gave an excellent description of the disease under the term leukochloroma. Since the myelocyte is the type of cell of the tumor, the term "myelocytoma" seems fitting. Myelocytoma is a neoplastic disease of myelocytes and may affect almost any tissue in the body.

*Histogenesis.* The myelocyte may be recognized readily as the type cell of myelocytoma, but the source of the tumor cells is an unsettled question. Cells which are morphologically similar to those of myelocytoma may be found in the bone marrow and in foci of extramedullary myelopoiesis of normal chickens. These represent normal metamyelocytes and myelocytes, which are immature acidophilic granulocytes. Two types of acidophilic granulocytes are found in the blood and the hemopoietic organs of the chicken. The more numerous are called heterophilic leukocytes and fulfill a function similar to that of heterophilic or neutrophilic leukocytes of mammals. The other type is the true eosinophilic leukocyte of mammals. Although ordinarily the adult heterophilic leukocyte of the chicken contains in its cytoplasm spindle-shaped acidophilic granules, the same granules are spherical at certain stages of their development. The granules of the eosino-

philic leukocyte are likewise spherical in the early stages of their development and remain so in the adult cell. Thus it is obvious that while the two types of leukocytes may be differentiated readily by the shape of the granules when adult in form, there are no reliable criteria for distinguishing between these cells in their immature stages of development. Although there are other features by which the two forms of adult cells may be separated, these features do not become apparent except in the later stages of development and are lacking in the type cell of myelocytoma.

During embryonal life, the mesenchyme in parts of the body other than the bone marrow acts as a hemopoietic tissue. This function subsides and at the time of hatching and afterward is almost entirely taken over by the bone marrow. The ability of tissue other than bone marrow to produce myelocytes is not entirely lost in post-natal existence. Foci of such cells developing in the periportal regions of the liver and in the thymus are encountered fairly frequently. The cells of a myelocytoma may arise from any or all of these potential sources.

The tendency for myelocytoma to involve many tissues or organs makes it difficult to determine whether the disease arises from a single primary focus or is an expression of a systemic disturbance. Mathews expressed the opinion that the disease was primary in the bone marrow and metastasized from there to the other sites in which the tumor was found. This position is hardly tenable in view of the possibility for the origin of myelocytoma in other tissues where a potential source of myelocytes exists. While it is possible that myelocytoma is a systemic disease in which numerous widely separated foci of neoplastic myelocytes have been developed as the result of an unknown stimulus acting simultaneously on multiple sources, such a conception is not entirely logical, for it would fail to explain why in some cases of myelocytoma the disease is confined to a single or to relatively few situations.

*Frequency.* Mathews (1929a) mentioned two flocks of chickens in which myelocytoma appeared as an enzootic. In one flock the losses from the disease were estimated to be 20 per cent and in the other flock 10 per cent. Although the flocks were small and not all birds that died were examined, this tendency of myelocytoma was well illustrated. Usually, however, myelocytoma is a sporadic disease in a flock. Thirty-six of the cases collected by Mathews were found during necropsy of 3,938 birds, an incidence of 0.91 per cent. Olson and Bullis (1942) found seventeen cases of the disease during the examination of 2,304 birds, an incidence of 0.74 per cent.

The age of birds affected with myelocytoma is usually less than one year. Mathews (1929a) found most of his cases during the winter months of November, December, and January but expressed the belief that the factor of age was responsible for this apparent seasonal effect. Olson and Bullis (1942) found the incidence of myelocytoma in each of the four quarters of the year to be the same when the factor of age was considered.

Mathews noted the disease to be common in chickens of the Barred Plymouth Rock breed. The data of Olson and Bullis suggest a greater frequency of myelocytoma in Barred Plymouth Rock chickens than in Rhode Island Red birds.

*Anatomic situations.* Study of the distribution of lesions of myelocytoma indicates that nearly any tissue or organ of the body may be affected with the tumor. A notable feature is the tendency for myelocytomas to develop on the surface of bones in intimate association with the periosteum. These may be sheetlike or nodular masses and frequently assume a peculiar, bilateral symmetrical aspect. Such bilateral symmetrical deposits often are found affecting the periosteum of the ventral portion of the keel bone and of the ribs. There seems to be a predisposition for the tumor to collect near cartilage at the costochondral junctions of the ribs and about the annular cartilaginous rings of the trachea. In some cases the tumor is disposed irregularly about the bodies of the vertebrae, especially in the lumbosacral region. Oberling and Guérin (1934b) described four such cases. Mathews found similar cases in which the tumor had infiltrated the bone, caused pressure on the spinal cord, and led to paralytic symptoms of transverse myelitis. Although myelocytoma tends to develop on the surface of the keel bones, ribs, vertebrae, and sometimes the flat bones of the skull, a similar involvement of the long bones of the legs and wings is infrequent.

The liver, spleen, ovary, and bone marrow, in addition to subperiosteal tissues, are affected with the neoplasm in most cases. Other organs and tissues are involved less frequently, although there is perhaps no tissue or organ which may be regarded as resistant to invasion by the tumor.

*Effects on the host.* Chickens coming to necropsy with myelocytoma are usually in a fair state of nutrition, suggesting either that the course of the disease is relatively rapid or that the disease has but little effect on the host. The former would appear more logical since the tumor itself has a distinctly malignant character and appears capable of rapid growth. In a few cases in which data on egg production were available, the period between cessation of egg production and death was short (average of 21 days in three cases). Mathews mentioned that the clinical symptoms of a slight indisposition observed in most cases did not exist for more than a week preceding death and that sudden death without noticeable symptoms sometimes occurred. Mathews also observed symptoms of transverse myelitis in two cases.

Relatively rare cases occur in which the tumor masses can be detected on examination of the exterior of the bird either about the head or about the sternum.

*Gross and microscopic description.* Myelocytoma has a characteristic appearance which is not likely to be confused with that of any other neoplastic tissue. The color is dull white. The tissue is soft and tends to be somewhat friable. In some instances the vascular bed of the tumor masses may be con-

gested, contributing a distinct pink cast to the color. An irregular infiltrative growth is typical of myelocytoma, and while localized masses may be found near bones, the growth in organs, as the liver, spleen, kidney, and lung, is usually diffuse. The liver, spleen, and kidneys, when affected with myelocytoma, become enlarged. However, the hypertrophy of these organs is not as marked as is commonly true in lymphocytoma.

The lesions of myelocytoma consist of infiltration with monotonously unvarying myelocytes. These cells, as previously mentioned, are similar to normal myelocytes found in the bone marrow and ectopic foci of myelopoiesis. Their nuclei are large, vesicular and usually eccentric in position and tend to be round or oval in outline, although often their shape is distorted by compression. A distinct nucleolus is commonly present. The cytoplasm is filled with acidophilic granules so tightly packed that their shape cannot often be distinguished, and although the granules are usually spherical, spindle-shaped granules may be noted in some cells. Imprint preparations made by touching a glass slide to the cut surface of fresh tumor material can be stained with a polychrome blood stain after drying in the air. Such a preparation may be compared with similar ones made of the blood or bone marrow. With such a stain the large nuclei have a very fine arrangement of the chromatin and parachromatin, and the cytoplasmic granules, while predominantly acidophilic, are occasionally basic in reaction. These basic staining granules represent a pre-acidophilic stage and later become acidophilic.

The myeloid tissue of the bone marrow becomes converted into a mass of tissue indistinguishable from foci of the tumor situated elsewhere. From the structure it must be considered as a neoplastic process in the marrow. The involvement of the bone marrow appears to be a constant feature and may occur in every case of myelocytoma. Mathews (1929a) and Olson and Bullis found the condition to exist in every case examined. In this respect, marked similarity exists between myelocytoma and granuloblastic leukosis. What appears to be a fundamental difference is that the neoplastic cells of myelocytoma are of a relatively later stage of development than those of granuloblastic leukosis. A careful comparative study of the minute structure of the bone marrow in myelocytoma and leukosis will reveal this difference.

Fairly frequently, abnormal myelocytes gain access to the blood stream. The morphologic characteristics of these cells are similar to those of the cells in the extravascular foci of tumor. A distinct increase of the number of heterophils in the blood may be noted also. Probably the extent of involvement of the blood will vary during the course of the disease, although this aspect has not been studied carefully.

*Metastasis and spread.* Although Mathews expressed the belief that myelocytoma is a primary tumor of the myeloid elements in the bone marrow, there are other reasons, as mentioned previously, to suggest that myelocytoma may develop simultaneously in several widely scattered regions.

The possibilities for such development have been discussed under the heading of histogenesis. When the process is once initiated, further development is infiltrative. Nerves, muscle, and bone may be invaded by the infiltrative growth of the tumor. Cartilage alone seems to be capable of resisting growth of the tumor, and this phenomenon may be studied readily when the trachea is involved. The presence of tumor cells in the general circulation provides a means of dissemination of the tumor; however, the importance of metastasis in the disease cannot be estimated.

*Special feature.* Mathews made unsuccessful attempts to transmit spontaneous myelocytoma. The Strain 2 agent developed by Furth (1933) has caused a neoplastic-like process, which he refers to as myelocytomatosis, in addition to lymphomatosis and endothelioma. The relation between this experimentally produced disease and spontaneous myelocytoma is not known. Nyfeldt (1934) reported development of a strain of leukosis in which leukemic myeloblastosis (granuloblastic leukosis) was the predominant type, although a few cases of aleukemic myeloblastosis (myelocytoma?) were also found in experimentally inoculated chickens. The occasional finding of myelocytoma as an enzootic in a given flock suggests the possibility of a common factor or factors as responsible for such an outbreak. For the present very little information is available on this point.

*Diagnostic characteristics.* Only brief comment is necessary to re-emphasize the comparative ease of recognizing myelocytoma. The color of the tumor and the distribution of lesions are features so characteristic of the disease that most cases of myelocytoma can be identified on macroscopic examination.

**Fowl leukosis.** Fowl leukosis is a disease of the myeloid tissues in which the precursors of erythrocytes and granulocytes are stimulated to unrestricted multiplication. The apparently functionless autonomous growth of myeloid tissue serves to characterize fowl leukosis as a neoplastic disease. The neoplastic character of the immature blood cells is also illustrated by their tendency to become immobilized within the vascular bed of certain organs such as the liver, spleen, and kidney. In these situations they display proliferative growth outside the confines of the bone marrow where under normal conditions they would ripen into mature blood cells before becoming released into the circulation. The tendency for immobilization has been called leukostasis.

The term "leukosis" has been used with a wide variety of interpretations in connection with avian diseases. It is used in this section in a restricted sense to indicate the single entity briefly characterized in the foregoing paragraph.[5]

---

[5] Other applications of the terms "leukosis" and "leukoses" are noted in the chapter on "The avian leukosis complex."

A thorough review of the extensive literature on fowl leukosis is beyond the scope of this section. Such a review was made in 1940 by Olson. The morbid anatomy of naturally acquired fowl leukosis is not different from that of the experimentally produced disease, and much of our knowledge has been gained from study of the experimentally produced forms of the disease.

*Histogenesis.* A consideration of the histogenesis of fowl leukosis must be based largely on evidence obtained from various experiments dealing with the transmissible forms of the disease. The stimulus responsible for the development of this condition is an ultramicroscopic agent present in the tissues of affected birds. Such an agent was demonstrated first by Ellermann and Bang in 1908. The agent of fowl leukosis can be demonstrated in a spontaneous case of the disease only by reproduction of fowl leukosis in other birds following the experimental introduction of the causative agent.

Although much work has been done with fowl leukosis, the site of inception and the mode of action of the causative agent remain unsettled. A review of the literature has revealed many facts and opinions with regard to the pathogenesis of this malady. Not all strains of the agent are similar. Some appear restricted in action and cause only one form of disease (such as erythroblastic leukosis), some may cause both erythroblastic and granuloblastic leukosis, and some may cause leukosis and fibrosarcoma. The production of either erythroblastic or granuloblastic leukosis by a single agent is not difficult to harmonize with the hypothesis that the agent may attack a stem cell (hemocytoblast) common to both cell lineages. The form of disease which develops apparently depends on either the reactivity potential of the affected stem cell or the ability of the agent to influence the line of differentiation of the stem cell.

The problem introduced by those agents of leukosis capable of also producing fibrosarcoma is more difficult to understand. The hemocytoblast and the fibroblast are related; yet this relation is somewhat distant, and it is rather difficult to believe that fibrosarcomas develop from hemocytoblasts stimulated by the agent of leukosis. The histogenesis of tumors produced by other agents such as the agent of the Rous sarcoma is not settled conclusively, although it is believed that the fixed or free histiocyte plays an important role in the process. Perhaps the agent responsible for leukosis may act in a similar way in the production of fibrosarcoma. Jármai (1935) explained the sarcoma-producing action of the agent of leukosis which he developed by suggesting that it had histotropic tendencies in addition to hemotropic tendencies, the latter being the more pronounced. Engelbreth-Holm and Rothe Meyer (1935) have advanced the conception that the different types of disease are caused by a selective action of the different agents of leukosis. For example, leukosis and fibrosarcoma are caused by an agent which attacks a

mesenchymal cell capable of forming either blood cells or fibroblasts; those agents causing either erythroblastic or granuloblastic leukosis attack a cell common to both; and those agents causing only erythroblastic leukosis attack a cell already committed to that cell lineage.

*Frequency.* Fowl leukosis is usually a sporadic disease among chickens and ordinarily affects birds more than six months of age. The average age of fowls suffering from leukosis studied by us has been approximately one year. Hamilton and Sawyer (1939) observed an unusual situation in which fifty-three of 231 chicks aged 30 and 39 days became affected with the disease within a period of two weeks. Olson and Bullis (1942) found seventeen cases among 2,304 chickens more than six weeks of age, an incidence of 0.74 per cent. Jármai (1934) expressed the belief that the disease has become increasingly prevalent from year to year, but precise information on this point is difficult to obtain.

Some evidence seems to suggest that in certain breeds (for example, Barred Plymouth Rock) of chickens, fowl leukosis is more likely to develop than in other breeds. Whether this is due to inherent characteristics of certain families within a breed or is a characteristic of all families of the breed is not known. Most if not all breeds are susceptible to transmissible strains of the agent of leukosis, and the spontaneous disease has been found in many different breeds.

There appears to be some relationship between the season of the year and the occurrence of the disease. Such relationship often has been suggested in the literature and is worthy of study. However, a number of factors serve to complicate such a study. In Denmark the disease appeared most often in the first quarter of the year; in Germany, during the autumn, winter, and spring; in Japan, in the late spring; and in Hungary, in the autumn and winter months. In Massachusetts it appeared most often in birds examined during the second quarter of the year. Engelbreth-Holm and Rothe Meyer (1932) noted a seasonal effect on the results following inoculation of chickens with the agent of leukosis. A more severe form of the disease was noted in the summer months, and the disease developed in a higher percentage of adult birds during April and May than during October and November. Jármai (1938) observed a longer interval between inoculation and death of experimentally inoculated birds in the first half of the year than in the last half.

*Gross and microscopic description.* Fowl leukosis is fundamentally a disease involving the myeloid tissue of the bone marrow. Pathologic changes in other organs or tissues are secondary to the basic process in the bone marrow. With this simple conception in mind the varied aspects of fowl leukosis as noted in other parts of the body may be understood readily.

The disease begins as a neoplastic proliferation of unripe erythrocytes or granulocytes. At first the process may resemble marked hyperplasia of

myeloid tissue, but soon the normal boundary lines between intravascular erythropoiesis and extravascular granulopoiesis are so disturbed that they can no longer be distinguished. The neoplastic blood cells gain access to the circulation and are released from the diseased marrow in ever-increasing numbers. The tendency for these cells to become lodged in the capillary bed of certain organs has been mentioned previously. In such regions of leukostasis, the neoplastic cells continue to multiply and may rupture the vessel wall and infiltrate the parenchyma of the organ or tissue. Sometimes the lumen of the vessel may be filled with leukotic cells to such an extent that they constitute a thrombus and lead to infarction of the region supplied by the blood vessel.

In fowl leukosis the bone marrow is grayish red and fills the marrow cavity. Johnson (1934) has called attention to the fact that in a normal bird the bone marrow space of the humerus contains fat and air spaces, whereas in most cases of leukosis the fat and air spaces are replaced by active myeloid tissue. A lining membrane of osteoid tissue may develop immediately inside the dense shaft of the long bones in cases with a protracted course. The intense hyperactivity of myeloid tissue can be studied best in histologic sections prepared from marrow of the long bones.[6] The marrow sinusoids are distended with unripe cells, and likewise, the intersinusoidal tissue conists of unripe granulocytes. The relative amount of each determines the type of leukosis. That is, in the erythroblastic form, erythropoiesis is more marked; and in the granuloblastic form, the intersinusoidal tissue is the more severely affected.

The liver, spleen, and kidneys are the visceral organs of predilection in which the leukotic blood cells tend to lodge and proliferate (Fig. 30.10). The proliferation leads to generalized enlargement of the affected organ. In general, this enlargement is not as marked as usually is seen in diffuse lymphocytoma. Microscopically, the leukotic cells are confined largely to the vascular bed. The parenchyma of the organ may be reduced by compression from distention of the vascular bed with masses of neoplastic cells. The color of the involved visceral organs is usually pale because of the anemia. Sometimes small white foci may be present and represent localized accumulations of leukotic cells.

Fowl leukosis is associated with a tendency to hemorrhage, probably the result of an early and marked reduction of number of circulating thrombocytes. Hemorrhages may be noted in the loose areolar tissues and in the mucosa of the intestine.

The circulating blood is affected in nearly all cases of fowl leukosis. In experimental leukosis cases may be observed in which the process is well

---

[6] The shaft may be split on its longitudinal axis to allow direct action of the fixative on the marrow. After fixation is complete a segment of marrow may be embedded in the usual manner.

developed in the bone marrow, and death occurs before the leukotic cells gain access to the circulation. Such cases are called incipient leukosis and represent a rapid acute form of the disease. All forms of immature erythrocytes and granulocytes may be found in the blood in varying numbers (Fig. 30.11). Usually the first change to be observed is a decrease of the number of thrombocytes. A decrease of the number of erythrocytes and of the amount of hemoglobin is followed closely or sometimes preceded by the appearance of immature cells. The blood picture is subjected to marked variations dur-
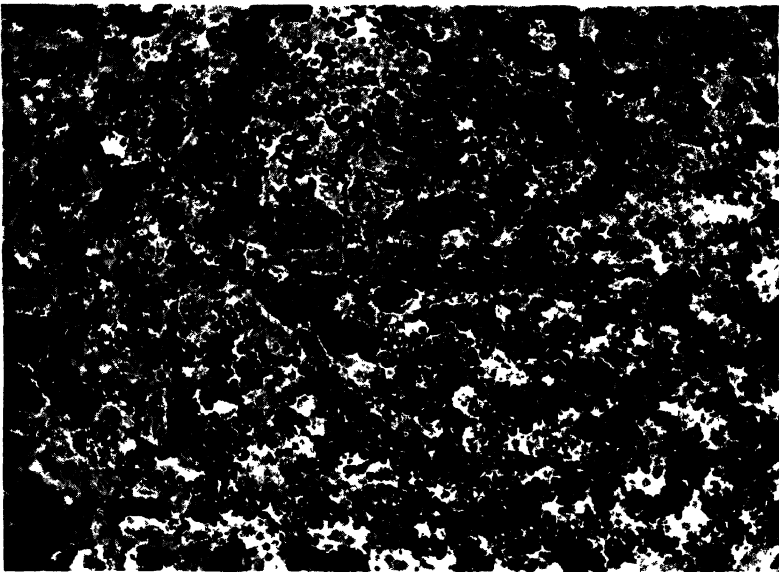


Fig. 30.10. Erythroleukosis showing marked engorgement of the capillaries of the liver by immature erythroblasts. ×195.

ing the course of fowl leukosis. The microscopic picture of granuloblastic leukosis may sometimes change to that of erythroblastic leukosis. In some instances the blood picture may become apparently normal and remain so, indicating recovery. Actual recovery of a spontaneous case has not been observed, although such a possibility may exist. In other cases periods of remission may be followed by the reappearance of pathologic cells in the blood, and the disease eventually proves fatal. Although the foregoing impressions were obtained from experimental data, comparable changes occur in the naturally acquired disease.

Many bizarre and unusual forms of blood cells may be seen in leukosis. Furth (1931) and Oberling and Guérin (1934a) have published excellent colored plates illustrating the various types of blood cells seen in the blood of chickens suffering from fowl leukosis.

*Special features.* The relative ease with which fowl leukosis may be transmitted by means of the agent of leukosis suggests that spontaneous cases may develop as a result of natural exposure to the causative agent. However, the contagiousness of fowl leukosis has yet to be established. Wickware (1946) found no evidence of transfer of leukosis to chicks hatched from pullets that had recovered from experimentally produced leukosis. Various experiments have indicated that not only fowl leukosis, but also the transmissible connective tissue tumors of chickens, are not contagious. Although
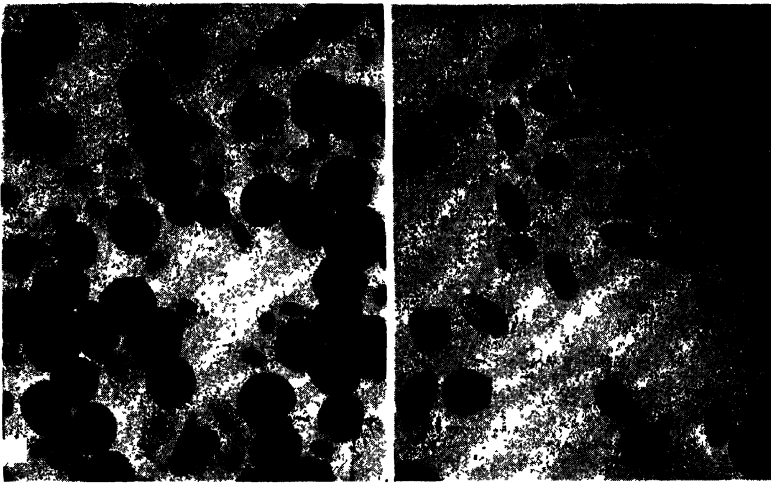


Fig. 30.11. Blood films of chickens affected with leukosis showing marked differences between the myeloid and the erythroblastic form of the disease. Left—myeloid leukosis. Right—erythroleukosis.

the agent of leukosis is present in all tissues of the body, the bile, urine, and feces are apparently not infective. Various ectoparasites have been shown capable of obtaining the agent from diseased birds and retaining it in an active form, but spread of the disease by such means can explain the development of few if any cases of leukosis. The spontaneous, endogenous origin within the host has been suggested for the agents of leukosis and transmissible connective tissue tumors. In this respect these agents would be entirely different from the filtrable viruses of contagious diseases such as fowlpox and laryngotracheitis. The concept of endogenous origin receives support in the different behavior of the many strains of the agent of leukosis, suggesting a lack of similarity. Tumors have been induced in chickens by carcinogenic chemicals, and if a tumor-producing agent separable from living cells could be demonstrated in these chemically induced growths, the evidence would support the hypothesis of endogenous origin of such agents. Only contradictory evidence is now available, and the latest report tends to deny the

existence of such an agent in artificially induced tumors (Murphy and Sturm, 1941a).

Fowl leukosis is a disease peculiar to chickens. Only one spontaneous case of the disease has been reported in another species of fowl. This case occurred in a small parakeet (*Melopsittacus undulans*) and was described by Jármai (1939). The specificity of action of the agent of leukosis is probably only relative since the disease has been produced experimentally in pheasants, turkeys, and guinea fowl.

Natural resistance to the agent of fowl leukosis may be found in some chickens experimentally inoculated, and those which recover from the experimental form of the disease also have a relative degree of immunity.

*Diagnostic characteristics.* The typical case of fowl leukosis is characterized by pale, watery blood which clots slowly, moderate enlargement of the liver and kidneys, marked enlargement of the spleen, and petechial hemorrhages in the loose areolar tissue and in the intestinal mucosa. The myeloid tissue fills the bone marrow space, replacing all fat cells and is gray red to dark red. Examination of smears of the blood will reveal abnormal numbers of immature cells.

In the diagnosis of leukosis, care must be exercised to differentiate it from other diseases such as secondary anemias, granulomatous processes, and other neoplastic diseases. Histologic examination of the myeloid tissue should be regarded as the basic requirement for the diagnosis of fowl leukosis, and in some cases this procedure must be supplemented by a microscopic study of other organs as well.

## MELANOMA

Melanoma is a pigmented tumor whose black color is due to the presence of melanin granules in the cytoplasm of the cells. Histologically, the melanin appears as fine, dustlike, yellow-brown particles which may become so concentrated as to obliterate entirely the structure of the cell. The pigment is produced by melanoblasts which produce an enzyme capable of transforming the colorless precursor of melanin into pigment. By means of the "dopa" (dihydroxyphenylalanine) reaction this enzyme can be detected and thus the melanoblasts identified. Melanin granules may be engulfed by phagocytes which become simply carriers (melanophores) of the pigment. The histogenesis of melanoblasts is unsettled, and according to the present conception, they may have either a mesodermal or a neural derivation (Boyd, 1938).

Excessive pigmentation with melanin without neoplasia may occur and is known as melanosis. Goldberg (1919) described a case of generalized melanosis in a turkey and cited a similar case observed by Lewin in a chicken. According to Kukleuski (cited by Reinhardt, 1930), pigmentation of the gonads, oviduct, thymus, thyroid, skin, and marrow is often

marked in Japanese and Siamese Silky chickens, which normally have a pigmented skin. Reinhardt (1930) commented that pigmentation of one or both testes is fairly common in "singing" birds. Melanosis of the peritoneum occasionally may be noted in the chicken.

Few pigmented tumors of chickens have been described. Reitsma (Hoogland, 1929) and Goldberg (1919) each reported a melanoma, probably primary, in the ovary of a hen, which spread to the serosa of the abdominal viscera. In the case described by Goldberg, the tumor resembled a cavernous angiosarcoma except for the pigmentation. McGowan (1928) described three cases of melanoma in the chicken. Two of the pigmented tumors occurred in Black Leghorn chickens and the other in a Rhode Island Red bird. In all of these cases the tumor was believed to have originated in the ovary. Only one appeared epithelial. The other two were described as similar to the Rous sarcoma. In McGowan's cases numerous implants of pigmented tumors were found on the serosal surfaces of the visceral organs. Olson and Bullis (1942) observed a small pigmented tumor at the base of the tongue which was diagnosed as melanoma. Ball (1945) reported a melanoma of the iris in a two-year-old hen that died with lymphomatosis.

## TUMORS OF NERVE TISSUE

If lymphocytoma is excluded, neoplasia of nerve tissue of the chicken would appear from the literature to be relatively infrequent. Jungherr and Wolf (1939) reviewed the literature on primary neural neoplasms of animals and found only three cases in the common fowl in which the diagnosis of glioma could be accepted. They described two additional cases. All were regarded as astrocytomas. Jungherr and Wolf also discussed neoplasms reported from other birds and regarded a glioma found in a parakeet and a ganglioneuroma described in a sparrow as of neural origin. They stated that the apparently low rate of incidence of neural tumors in animals is perhaps due to the infrequent complete examination of the central nervous system at necropsy.

Jackson (1936a) has described multiple neurofibromatosis in the chicken, and Olson and Bullis reported five cases of neurogenic sarcoma. These tumors are mentioned in the section dealing with connective tissue tumors. Cole in 1946 reported a case of retinoblastoma.

No attempt will be made to discuss the classification or characteristic features of tumors of the nervous system. Those seeking such information are referred to the chapters on neoplasms in Penfield's (1932) handbook.

In some instances the brain or spinal cord may be affected by metastatic growth of neoplasms situated elsewhere. The involvement of nerves with lesions of lymphocytoma is fairly frequent in that disease. The significance

of these lesions and their association with fowl paralysis are discussed in the section on lymphocytoma.

## TUMORS OF EPITHELIAL TISSUES

**Papilloma.** A palilloma is a benign epithelial tumor composed of fibrous cores or projections which are covered by layers of epithelial cells. These tumors are frequently multiple, and the brushlike or cauliflower-like structures are often spiny to the touch.

Microscopically, papilloma is a simple structure usually consisting of a few to many separate units or projections, each with a fibrous core that is covered to a variable depth by compactly arranged epithelial cells. The cells nearest the stroma are the least mature, and between the various papillae and on the surface there is frequently present a horny deposit known as keratin. Keratin is the product of the more mature squamous epithelial cells. Characteristically, these tumors grow in an outward rather than an inward direction.

Our experience appears to be in agreement with that of others concerning the frequency of occurrence of papilloma in chickens. The tumors are sometimes seen on the comb, feet, and wattles of fowls, but they occur much less frequently in fowls than in certain mammals. Olson and Bullis (1942) observed a case of multiple papillomatosis of the esophagus of a chicken. The lesions appeared as small, grayish nodules, some of which were hemorrhagic, in the mucosa. We observed one instance in a pigeon in which there occurred diffuse warty growths in the skin adjacent to the beak and around the eyes. The literature contains but slight mention of papillomas in chickens, and one must conclude that their occurrence is extremely infrequent.

Papilloma of the skin and oral cavity in mammals frequently is contagious and easily transmitted owing to a causative factor that in some instances has been definitely established to be a virus. So far as we know, papillomas of chickens due to agents of a virus-like nature have not been reported. Taken in all, papilloma insofar as chickens are concerned occurs so infrequently as to be of little importance.

**Adenoma.** Adenoma may be defined as a benign epithelial neoplasm in which the structural pattern resembles that of a gland. Any tissue containing glandlike structures normally or aberrently may give rise to an adenoma. These tumors usually occur singly. Rarely, multiple adenomas may appear.

Adenomas are among the less frequent tumors of chickens, being much less common than the malignant epithelial tumors. The largest series of adenoma of chickens with which we are familiar is that encountered by Eber and Malke (1932). These authors reported the occurrence of sixteen ade-

nomas among 253 tumors of fowls. Of those in chickens, the sites of occurrence were as follows: liver, six cases; proventriculus, one case; gizzard, one case; intestines, two cases; ovary, one case; and oviduct, one case. Eber and Malke also recorded two cases of adenoma of the kidney. However, the most frequent neoplasm of the kidneys of chickens is embryonal nephroma, and it is possible that the two cases of adenoma of this organ mentioned by Eber and Malke were in reality embryonal nephromas. According to Heim (1931), Joest and Ernesti described a cystic form of adenoma (cystadenoma) in the region of the crop. Although the exact situation of origin was not determined definitely, origin from the thyroid was considered. A few cases of adenoma of the ovary have also been described (Heim). Olson and Bullis observed one case of fetal adenoma of the thyroid and one case of adenoma of the feather matrix, in chickens. The same authors recorded a papillary cystadenoma of the mucosa of the posterior portion of the gizzard in a chicken.

The abundance of normal tissues that contain epithelial glandlike structures provides in chickens potential sites for the occurrence of many more adenomas than are encountered. Apparently in the chicken when epithelial glandlike structures undergo autonomous changes, the resultant neoplasm is more likely to be malignant than benign.

Unless adenoma occurs in a situation where its presence or size may provide a mechanical interference with the proper functioning of contiguous tissues, this variety of neoplasm in chickens is unlikely to have any appreciable effect on the host. Malke, according to Heim (1931), recorded an obstructing adenoma of the cecum. Babic (1931) described multiple adenomatous polyposis of the intestine in a chicken. Such a condition could lead to disturbances of elimination. Should adenoma arise in certain endocrine structures, abnormal physiologic effects may ensue.

Being benign, adenoma never infiltrates the surrounding tissues and does not metastasize. Should an alleged adenoma exhibit these features and especially should metastasis occur, the tumor can no longer be considered benign but should be recognized as malignant. The malignant form of adenoma is designated "adenocarcinoma."

In appearance adenomas may be expected to be encapsulated, nodular, and firm to soft swellings. Opportunity for adenoma to become cystic is provided by the glandlike nature of the parenchyma. Since a duct system for the natural egress of secretory substances is missing, the products of the cells frequently accumulate and produce small to large cysts. Such tumors are often called "cystadenomas."

Microscopically, an adenoma presents the appearance of a gland, a duct or a tubular structure (Fig. 30.12). Alveolar spaces may be present, or the parenchymal cells may appear as compact masses. In nearly every instance

the structure of the tumor bears a resemblance to the normal tissue produced by the parent epithelial cells from which the parenchymal cells of the tumor were derived. The stroma consists of fibrous connective tissue in which are found blood vessels. The stroma may occur in a promiscuous, nondescript fashion, or it may be disposed as septa or ill-defined trabeculae which serve to separate the tumor into irregular lobules.

Adenomas may be identified readily if one keeps in mind certain salient features: 1. Adenomas originate in situations where glandlike structures
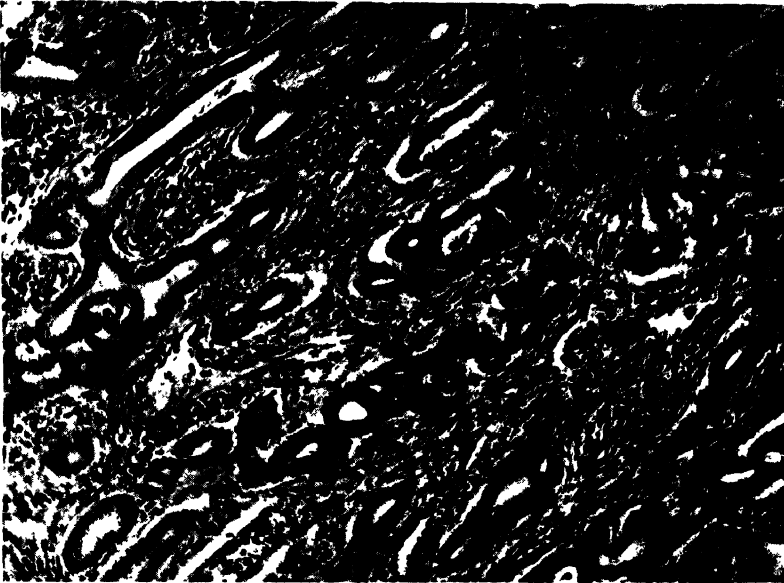


Fig. 30.12. Adenoma arising from the bile ducts of the liver. ×120.

occur normally. 2. They usually occur as single tumors. 3. They do not infiltrate the adjacent tissues or set up distant metastatic growths. 4. When properly removed they do not recur.

**Carcinoma.** A carcinoma is a malignant neoplasm composed of epithelial cells and a stroma of connective tissue. The latter provides a supportive structure for the epithelial cells and for the vascular channels inherent to the part. The cells of the carcinoma proliferate in an atypical and lawless manner, have a tendency to infiltrate and destroy the contiguous tissues, and may and often do set up secondary or metastatic foci. Although all malignant tumors of which the type cell is epithelial in origin are properly referred to as carcinomas, certain characteristic structural differences occur that make it desirable to separate carcinoma into several distinct morphologic types. These include adenocarcinoma, in which the parenchymatous cells assume a glandular or ductlike arrangement; squamous cell carcinoma, composed of

diffuse masses or compact collections of cells that arise from the epidermis or the mucosa of the esophagus, the mouth, or the pharynx; papillary carcinoma, which has a rough cauliflower-like surface with the tumor cells arranged in finger-like sheets; and hepatic cell carcinoma, which arises from the cells of the parenchyma of the liver which have undergone autonomous transformation. Tumors of chickens analogous to the so-called basal cell carcinoma of human beings have not been reported heretofore. Olson and Bullis (1942) encountered one case in their material. This is described in the subsequent text. Other special types of carcinoma may arise from the thyroid, the adrenal, the ovary, the kidney, and the pancreas. These constitute only a partial list of the tissues that may give rise to carcinoma. The wide distribution of epithelial tissues throughout the body provides numerous potential situations for the origin of carcinomas.

*Frequency.* In attempting to ascertain the approximate frequency of occurrence of malignant epithelial tumors in chickens, one is confronted with difficulties. First is the paucity of statistical data based on a sufficiently large amount of material to be significant and in which the diagnosis has been established by the microscopic examination of the tissues. Furthermore, the question of what should or should not be considered as neoplasia has an important bearing on the relative incidence of the various types of tumors.

Among 199 neoplasms of chickens mentioned by Hoogland (1929), thirty-three, or 16.6 per cent, were carcinomas. The predominance of sarcoma over carcinoma was well illustrated in Hoogland's series, there being ninety-three tumors of sarcomatous character. Hoogland's list of chicken tumors did not include those of so-called leukotic character (lymphocytoma, myelocytoma, and leukosis). Eber and Malke (1932) observed twenty-nine carcinomas among a total of 239 tumors of chickens. This represents an incidence of approximately 12 per cent. As was true in Hoogland's series, Eber and Malke probably did not include leukosis in listing the respective neoplastic diseases in their material. However, it is likely that many, if not all, of the so-called round-cell sarcomas mentioned by Eber and Malke were in reality lymphocytomas. As is true with most other reports on neoplasia of chickens, Eber and Malke's material contained many more "sarcomas" than carcinomas. Of the 239 tumors of chickens, examined histologically, 167 or approximately 70 per cent, were designated as sarcoma.

Babic (1931) described sixteen cases of carcinoma of the chicken, in ten of which the tumor was primary in the ovary. In three the tumor was primary in the skin, in two the kidney was the site of origin, and in one instance the growth arose in the testes. The tumors occurred in a group of sixty-one neoplasms collected from several different species of birds.

Goss (1940b), who reported on the types of neoplasia among 7,408 chickens examined at necropsy, found tumors in 1,445. Among 1,104

examined microscopically there were 991 designated "leukotic tumors" and seventy-seven carcinomas. In relation to the total number of tumors listed (1,104), carcinomas represented 7.0 per cent; if the "leukotic tumors" are excluded from the total number, carcinomas represent approximately 68 per cent of the neoplasms examined. Goss's data are of especial interest because of the fact that seventy, or approximately 91 per cent, of the total number of carcinomas found were ovarian in origin.

Olson and Bullis (1942) made a study of avian neoplastic material that occurred in 365 chickens. A total of 384 tumors, including those of hemo-blastic origin, were found, and twenty-four, or approximately 6.2 per cent, were epithelioblastomas.

From the figures presented it is evident that dependable information concerning the predictable rate of occurrence of carcinoma in chickens is meager. Many factors contribute to the unsatisfactory state of our knowledge concerning this question. Among these may be mentioned (1) failure to obtain in many instances a microscopic diagnosis of chicken tumors; (2) the wide diversity of origin of chickens submitted to most diagnostic labora-tories; (3) lack of important information concerning many of the flocks from which chickens suffering from neoplastic conditions are obtained; and (4) failure to have examined at necropsy by a competent pathologist, the bodies of all chickens dying of whatever cause, this examination to apply also to the carcasses of all chickens intended for food.

*Relation to age.* There do not exist adequate data to enable one to state definitely the relation of the incidence of carcinoma to the age of the affected bird. Indications are that the majority of carcinomas of chickens occur in adult rather than in young birds. Most birds that have carcinoma are one year or more of age. It should be kept in mind, however, that the age at which the neoplastic process began was much earlier.

*Sites of occurrence.*[7] As mentioned previously carcinomas may arise wherever epithelial tissues occur. Although the epidermis and the mucous membranes constitute the greatest amount of epithelial tissue in the body, in the chicken these tissues do not give rise to the largest number of carcinomas. In chickens most tumors of this character arise from the ovary.

From information obtained from the literature and from data supplied by our own collection, the occurrence of carcinoma in the various anatomic situations in chickens will be described briefly.

1. *Integument.* Although the only specimens of carcinoma of the skin in our collection occurred on the neck and head, by far the greatest number of carcinomas of the integument of chickens that have been reported have af-fected the foot and shank or more specifically the skin overlying the meta-

---

[7] A fairly complete résumé of the literature pertaining to carcinoma of chickens is to be found in the treatise by Reis and Nóbrega (1936).

tarsus.[8] Other situations in which carcinoma of the skin has been reported include the anal region, breast, and the region overlying the femorotibial articulation.

One instance of a tumor that had many of the characteristics of a basal cell carcinoma was reported by Olson and Bullis (1942). The tumor was situated in the skin immediately over the left eye. It was a nodular mass 1 cm. in diameter by 6 mm. thick. The tumor was first observed one month before the chicken was killed for necropsy. Microscopically, the mass consisted of several indistinctly lobulated, compactly disposed epithelial cells with moderately basophilic cytoplasm. The tumor was situated largely in the corium but had broken through the epidermis at one point. The structure was richly vascular, and some hemorrhage had occurred. Mitosis was not observed. Metastasis had not occurred. The tumor was diagnosed as carcinoma of the feather matrix.[9]

In view of the fact that these tumors of the integument have all the morphologic characteristics of a malignant growth, it is somewhat surprising that they apparently seldom if ever metastasize (Fig. 30.13). Duran-Reynals (1946b) was unable to transplant a localized skin gland adenoma of the wing to other chickens. In the cases we have observed, the neoplasms have remained localized, and in the cases reported previously by others, metastasis has rarely been demonstrated. Structurally, these tumors would appear capable of setting up metastatic foci at a distance from the original lesion. Yet in the many cases reviewed by Abels (1929) metastasis was recorded in only three cases of carcinoma of the skin of chickens.

2. *Alimentary canal.* A few instances have been recorded of the occurrence of carcinoma within the oral cavity of chickens (Heim, 1931, listed several cases). One occurred in our collection. The tumor was a carcinoma of the epidermoid type and occurred in the pharynx of a two-year-old hen. Although the tumor was locally infiltrative and destructive, metastasis could not be demonstrated. Incidentally, this tumor was responsible for considerable distress when the hen breathed. When breathing, the hen was said to be "gasping for breath."

The literature, according to Heim (1931), yields three cases of squamous cell carcinoma of the esophagus.

Carcinoma of the proventriculus and gizzard appears to be an extremely rare manifestation of the disease. Our material did not contain any specimens from these organs, and the cases reported in the literature are few. Babic (1931) described a medullary carcinoma of the proventriculus. Zannini

---

[8] The two specimens from the skin of the neck were received many years ago from Dr. F. P. Mathews. Both tumors occurred in chickens of the Barred Plymouth Rock breed, and each chicken was approximately one year of age.

[9] For a review of the different reports of carcinoma of the skin of chickens up to 1930, the paper by Heim (1931) may be consulted. Other cases are mentioned by Pohl (1926), by Babic (1931), and by Jackson (1936, page 434). An extensive review of the literature pertaining to tumors of the skin of birds will be found in the paper by Abels (1929).

(Heim, 1931) is said to have observed an adenocarcinoma of both the proventriculus and the gizzard. "Cylindrical cell" carcinomas of the gizzard that had not metastasized were reported by Schöppler (1913) and by Prospero (Heim, 1931). The report of Prospero was not available for review, so that we are unable to comment concerning this case. In Schöppler's case the tumor was considered to have arisen from the glandular elements of the pyloric portion of the gizzard.

3. *Intestine.* The tendency of many primary malignant tumors of the abdomen to spread by direct continuity or by serosal implantation to all of
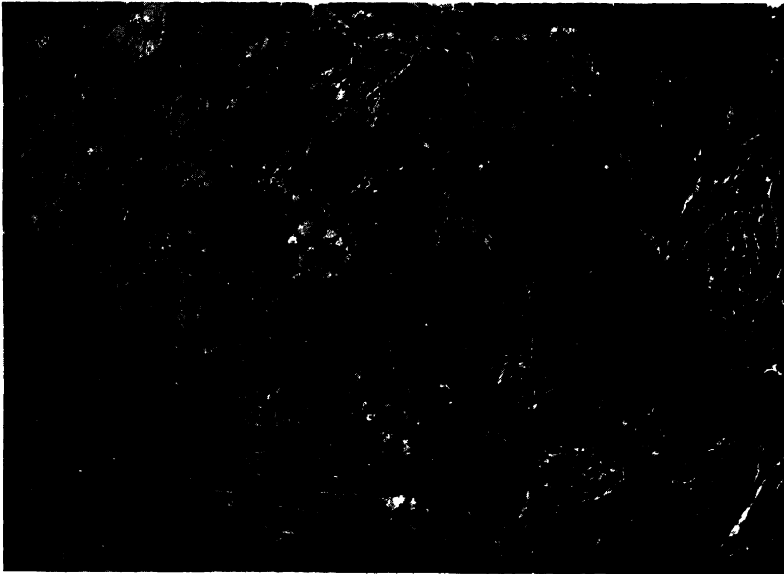


Fig. 30.13. Squamous carcinoma of the skin of the neck of a chicken. Metastasis had not occurred. ×120.

the serous surfaces of the abdomen frequently makes it difficult to ascertain with certainty from what site a given tumor may have arisen. The occurrence of serosal implantations is especially characteristic of ovarian carcinoma, and the resultant widespread distribution of the tumorous tissue may obscure entirely the primary site of origin or lead to false conclusions regarding the primary situation of the tumor. Unless one can demonstrate the primary lesion in the intestinal mucosa, it would be unwise to claim that a carcinoma of the intestine is present. When the serous covering or even the muscle wall of the intestines is involved with an epithelial glandular type of malignant lesion, discretion should be exercised in concluding that the site of origin was the intestinal mucosa. As a matter of fact the likelihood that such tumors originate in the ovary is much greater than that of their origin in the intestine.

Adenocarcinoma of the duodenum has been reported by Petit and Germain (Pohl, 1926) and by Ehrenreich and Michaelis (1906)[10]; and of the "intestine" by Hoogland (1929) and by Jackson (1936, p. 160). In Jackson's case metastasis to the liver had occurred. An unusual case was that of Joest and Ernesti (Heim, 1931), in which a medullary carcinoma involving the ileum and ceca was associated with another primary carcinoma of the cloaca. One of the specimens in our collection was of some interest. The tumor, which proved to be an adenocarcinoma, occurred in the mucosa of the ileocecal junction of a two-year-old White Leghorn hen (Fig. 30.14). The
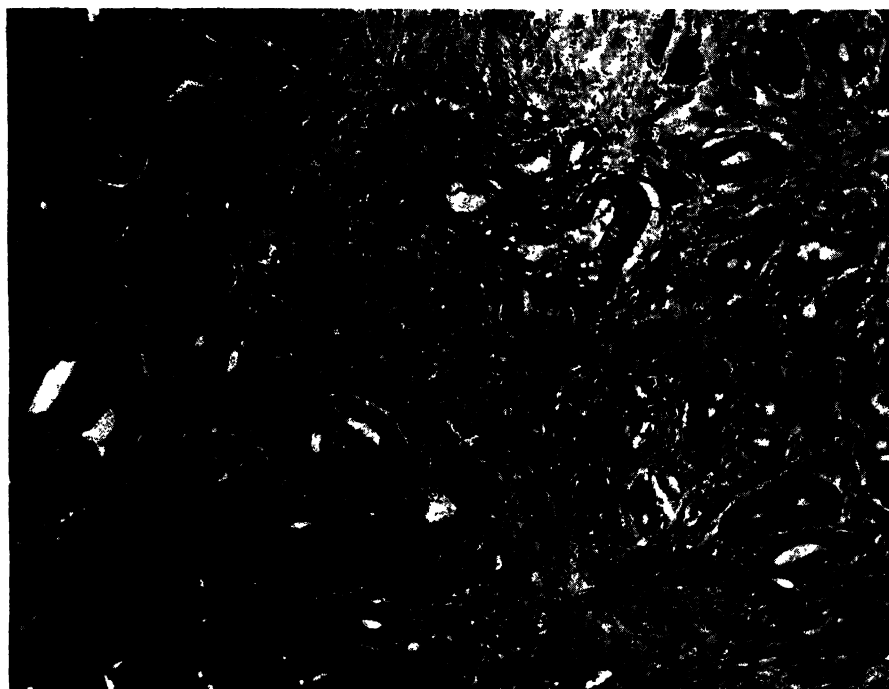


FIG. 30.14. Primary adenocarcinoma of the ileocecal junction of a two-year-old White Leghorn hen. The advancing neoplastic cells have penetrated the muscularis mucosae. Distant metastasis was not demonstrated. ×110.

tumor was an elongated, roll-like structure about 0.6 cm. in height, and it involved about half of the circumference of the lumen. The tumor had produced some obstruction, but metastasis had not occurred.

According to Heim (1931) two cases of carcinoma of the colon or rectum of chickens have been described. In both instances metastasis had occurred to the liver.

---

[10] Of the two cases of intestinal carcinoma reported by Ehrenreich and Michaelis, the facts presented in one failed to establish definitely that the tumor had arisen from the duodenum and not the oviduct.

The evidence indicates (1) that malignant epithelial tumors of the intestines of chickens are among the rarer avian neoplasms; (2) that carcinoma of the intestines may metastasize to the liver and lungs; (3) that in female chickens the occurrence of multiple neoplastic foci on the serosa of the abdomen should suggest a malignant lesion of ovarian origin rather than one from the intestinal tract; and (4) that to diagnose with certainty a carcinoma of the intestine, one should demonstrate the primary lesion in the mucosa of the gut.

4. *Accessory organs of digestion.* Among the accessory organs of digestion that have given rise occasionally to carcinoma are the liver and the pancreas.

Heim listed a few reports of epithelial tumors of the liver, but how many of the alleged cases were actually primary carcinomas of the liver is uncertain. Teutschlaender (Heim, 1931) reported two cases of carcinoma of the bile ducts, and Savage (1926) reported the occurrence of an adenocarcinoma of the gall bladder in a three-and-a-half-year-old Rhode Island Red hen.

Goss reported one case of carcinoma of the liver cells in a bird more than three years old and also an adenoma of liver cells. He also recorded four carcinomas of bile ducts. Olson and Bullis (1942) described three cases of benign hepatoma and four of cholangioma. In three instances the tumors of the bile duct cells were single isolated masses, and in the fourth case there were multiple tumors scattered throughout the liver, suggesting the malignant character of the neoplasm. An adenocarcinoma primary in the liver of a turkey was described by Babic (1931).

In the consideration of epithelial tumors of the liver, one should keep in mind, as Jackson (1936, p. 137) suggested, that it may be difficult to distinguish nodular hyperplasia, adenoma, and carcinoma. Primary carcinoma of the liver may arise from two types of cells, the parenchymal liver cells and the epithelium of the bile ducts (Figs. 30.12 and 30.15).

Olson and Bullis (1942) recorded having encountered three epithelial malignant lesions of the pancreas. Two of these were designated as carcinomas, and one was listed as an adenocarcinoma. Babic (1931) also described an adenocarcinoma of the pancreas.

5. *Adrenal glands.* Mathews and Walkey (1930) described in adult hens six cases of pedunculated carcinoma which appeared to originate from the region of the adrenal glands. Although the ovary was involved in each instance, the histologic picture of the tumors was suggestive of the adrenal cortex. Mathews and Walkey, however, were not certain that the tumors had originated from this tissue. Metastasis was limited ,to the mesentery and to the visceral peritoneum and probably occurred as a result of spread by continuity.

Berner (1923) reported an extremely interesting case of hypernephroma or carcinoma of the right adrenal gland with secondary involvement of the

serosa of the abdominal viscera and metastatic foci in the lungs. The bird was an eighteen-month-old female chicken in which the behavior and other characteristics of virilism had been noted since the bird was six months of age. There had developed a male type of comb and spurs, and the gait when walking was particularly vigorous and malelike.

6. *Urogenital tract.* A case of medullary carcinoma of the kidney with metastasis to the liver and lung was found in a pheasant by Babic. He also reported a cystic adenocarcinoma in the right kidney of a chicken. The latter case might be considered more properly as an embryonal nephroma. A medullary carcinoma of the testis of a chicken with metaplastic keratinization also has been described by Babic. Otherwise, we do not know of any instance in which the diagnosis of carcinoma of the testes has been established definitely.[11]



FIG. 30.15. Hepatocellular carcinoma (hepatoma). The rows of neoplastic cells and the capillary nature of the stroma are characteristic. ×300.

The ovary is undoubtedly the most frequent site of origin of carcinoma of the chicken.[12] In Eber and Malke's series of 239 tumors of chickens, twenty-nine were carcinomas. Fifteen, or approximately 52 per cent, of the carcinomas in this series were presumed to have originated in the ovary. Unlike many carcinomas of the ovary of human beings which affect the ovary secondarily, most carcinomas of the ovary of chickens are primary in this organ. The disease in chickens usually has its inception in the functioning left ovary but may originate in the rudimentary right ovary.

Seifried (1923) described two cases of ovarian tumors in chickens which he designated "oophoroma folliculare." One tumor occurred in the rudimentary right ovary and the other in a left ovary which was otherwise normal.

---

[11] Pohl described what he designated as an adenocarcinoma of the testes of a rooster. The tumor was of variable structure and contained cystic cavities and regions of myxomatous degeneration. Pohl's description of the tumor makes the diagnosis of adenocarcinoma unconvincing.

[12] Carcinoma of the ovary of chickens has been described by many observers. Those especially interested may consult the following: Oshima and Roki (1925), Eber and Kriegbaum (1916), and Jackson (1936b). Heim reviewed the literature up to 1930.

From a careful reading of Seifried's description of the two tumors, we believe that his Case 1 represents a granulosa tumor and not a Brenner tumor as he believed. Case 2 may also be a granulosa tumor, but the evidence is somewhat indefinite.

In the ovary, neoplasia may arise from (1) the germinal epithelium, (2) the Pflüger egg cords, and (3) the immature follicular epithelium.[13] Babic expressed the belief that most ovarian carcinomas are derived from follicular epithelium. Carcinomas that arise in the oviduct are derived from the epithelium of the mucosa.

Notwithstanding the possibilities for origin of ovarian carcinoma just listed, it should be kept in mind that, with the possible exception to be mentioned, cytologic detail of carcinoma of the ovary does not bear any resemblance to any recognizable ovarian structure. A possible exception to this statement is to be found in the case of granulosa tumor of the ovary. Even with this neoplasm, however, there is little unanimity of opinion concerning its true genesis. Most carcinomas of the ovary are medullary in character.

Attempted transplantation of an adenocarcinoma of the ovary to other chickens by Duran-Reynals (1946b) was not successful.

7. *Other situations.* Information on the occurrence of primary carcinoma of the lung of chickens indicates that this organ is seldom affected. The only reference to primary carcinoma of the lung of the domestic chicken with which we are familiar is a report of a case by Apperly (1935). The right lung and most of the left lung were replaced by a fibrocaseous material, and there was present a small white nodule near the periphery of the liver. Microscopically, the appearance of the lesions in the lung and in the liver was essentially identical. The diagnosis was primary adenocarcinoma of the lung with metastasis to the liver.

Epithelial malignant lesions of the thyroid gland appear to be rare in the chicken. Elsner (Reis and Nóbrega, 1936) described an adenocarcinoma, and Olson and Bullis a fetal adenoma in the thyroid of chickens.

*Effects on the host.* The effects on the welfare of an animal in which carcinoma develops depend on several factors. These include (1) the inherent malignancy of the process, (2) the anatomic situation involved and the severity of the involvement, (3) the presence or absence of secondary foci, and (4) the duration of the disease.

Carcinomas of the ovary frequently spread by continuity or by implantation to the adjacent structures and especially to the serosa overlying the intestines. When this occurs moderate to severe ascites eventually follows.

---

[13] Fischel (1922) stated that most of the ovarian tumors in human beings are derived from primitive mesenchymal tissue which has divergent potentialities, and which in his opinion may give rise to sarcoma or to carcinoma.

As a matter of fact the presence of ascites in an adult chicken should suggest the possibility of abdominal carcinomatosis or other malignant processes having a multiplicity of lesions.

It is of interest that although carcinoma of the abdominal cavity may be disseminated extensively throughout the viscera, frequently the condition appears not to interfere seriously with the general health of the bird except in the terminal stages. The disease has a protracted course, and in many instances is discovered in apparently healthy chickens at the time the bird is being "dressed" for food. Emaciation may be noted.

Hens that have carcinoma of the ovary eventually become nonproductive, although what degree of involvement of the ovary must occur before egg production ceases is not known.

Although the effect of most carcinomas, like that of many other malignant tumors, depends on the mechanical interference with normal function and on the destruction of normal tissues by the advancing neoplastic process, a few carcinomas occur that affect the host physiologically by producing an excess of hormonal substances. The masculinizing effect of arrhenoblastoma in the human being is well recognized, and if this or certain other specialized tumors occur in chickens it would seem reasonable to expect an altered behavior on the part of the host.[14]

*Gross characteristics.* The gross appearance of carcinomas varies greatly as to size, shape, and color. With few exceptions these tumors are attached diffusely to the surrounding tissues, and encapsulation except in rare instances is not discernible. Contact with the adjacent tissues is usually intimate, this feature being indicative of the characteristic invasive tendencies of these neoplasms. Those on the exterior, being subject to trauma, are readily infected, and as a consequence they may present hemorrhagic, necrotic regions. Their surface is usually irregularly roughened and may have a hard, tough, or cornified superficial covering due to the excessive amount of keratin derived from the squamous epithelial cells.

Carcinomas of the internal organs, especially those of the ovary or oviduct, are single or more frequently multiple, smooth, nodular, pinkish-gray or flesh-colored formations (Fig. 30.16). Multiplicity is frequently a striking feature of intra-abdominal carcinomas, with innumerable nodules of varying sizes and shapes occurring in subserosal situations, especially along the intestines and in the mesentery (Fig. 30.17). Ovarian and oviductal carcinomas may be soft or hard or, more precisely, cystic or solid. Cavities or cysts filled with a shiny mucinous or gelatinous substance are commonly a characteristic feature of these tumors.

---

[14] Friedgood and Uotila (1941) described several cases of virilism in chickens apparently due to hormones produced by tissue which they considered neoplastic. They diagnosed these cases as arrhenoblastomas. Virilism in a hen should lead to suspicion of such a condition.

Carcinomas of the mucous surfaces are often the site of extensive ulceration. The surface may appear raw and irregular with evidence of recent bleeding. Signs of secondary infection are present. In situations where metastasis has occurred, as in the lung or liver, the tumorous tissue usually appears as firm, grayish-white foci. In the liver the metastatic foci may be limited to the serosa of the capsule or may occur firmly embedded in the parenchymal tissue.

*Microscopic characteristics.* Since carcinomas arise from many different types of epithelial tissue, the histologic pattern depends to some extent on



Fig. 30.16. Solid carcinoma of the ovary.

the character of the parent epithelium from which they originate. However, most carcinomas have one important feature in common. This feature is the infiltrative and destructive character of their growth.

The various types of carcinoma that may be encountered can be described briefly as follows:

1. *Squamous cell carcinoma.* These tumors arise from the cells of the epidermis and consist of diffuse collections or compact masses of squamous epithelial cells. The epithelial cells are usually incompletely separated by a fibrous connective tissue stroma with nestlike groups or packets or finger-like processes that extend into the surrounding nontumorous tissue. Many if not most of the epithelial cells show signs of immaturity, and mitotic division usually can be seen. Necrosis may be present, and lymphocytes and

acidophilic granulocytes commonly infiltrate the tumor. The central portion of many of the compact groups of cells frequently presents a peculiar hyaline appearance due to the accumulation of keratin. Such a tumor may be referred to as a "pearl cell" or an epidermoid carcinoma.

2. *Papillary carcinoma.* Rarely a carcinoma grows away from, rather than into or toward, the body, presenting a rough, cauliflower-like structure. Such a tumor consists of a central core of connective tissue from which are given off small papillae covered with several irregular layers of neoplastic epithelial cells.



FIG. 30.17. Adenocarcinoma of the ovary showing multiple implants on the serosa of the oviduct of the mesosalpinx.

3. *Adenocarcinoma.* This variety of carcinoma arises from tissues normally composed of or containing epithelial glandular structures. The tumor consists of fibrous stroma of variable dimensions which supports one or more layers of cuboidal or columnar cells arranged in an alveolar or ductlike fashion (Fig. 30.18). Tortuous ductlike structures frequently are formed, as are cystic spaces containing gelatinous or mucoid material. The cells may assume a papillary type of growth, and if the process is especially anaplastic little tendency toward formation of alveoli or ducts may be evident. Instead, the growth appears as diffuse or solid sheets or nests of cells with slight resemblance to the parent structure from which the tumor arose.

4. *Other types of carcinoma.* The paucity of information concerning carcinomas derived from the hepatic cells of chickens indicates the infrequent

occurrence of this variety of neoplasm. In the one case described by Jackson (1936a) the essential histologic features may be summarized as follows:

The structure was without signs of lobulation, a collagenous stroma was absent, and bile ducts did not occur. There were present rather large, irregularly scattered blood vessels. The stroma was purely capillary in character and served to separate tubular, cordlike arrangements of cells significantly like those of the hepatic parenchyma. A predominant feature was the glandlike or acinous appearance of the neoplastic process. Distinct formation of lumina by the epithelial cells was readily discernible under
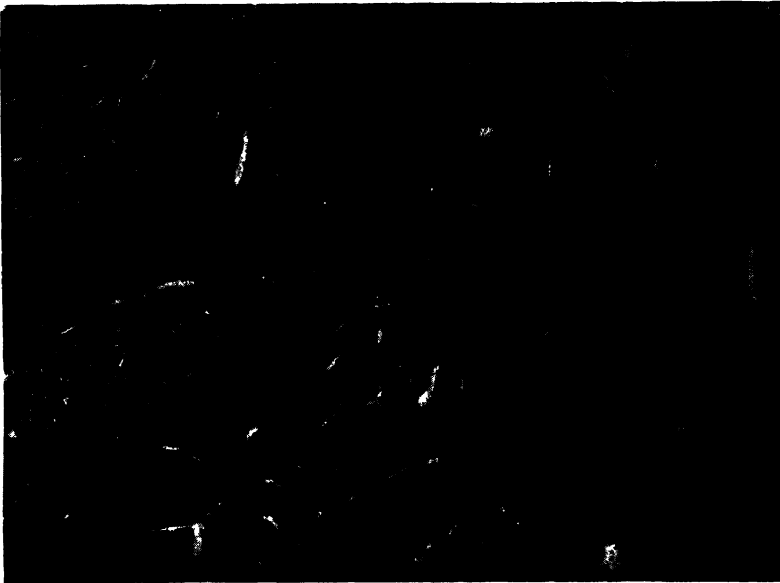


Fig. 30.18. Cystadenocarcinoma of the ovary of a chicken. Metastasis to the lungs had occurred. ×150.

low magnification (see Fig. 30.15). A malignant neoplasm arising from the hepatic cells is properly designated "carcinoma hepatocellulare."

Another variety of epithelial malignant lesion is that commonly referred to as hypernephroma of the kidney. This is a renal carcinoma that arises either from the adult tubules or from foci of nephrogenic tissue that have persisted in the renal cortex. Hypernephroma of the kidney appears to be rare in chickens.

Recently one of us encountered a hypernephroma in the kidney of a hen about one year old. The bird died with extensive lymphocytoma of the visceral organs. The hypernephroma was a well-encapsulated, firm, round mass in the posterior pole of the left kidney. The tumor was 4.5 cm. in diameter. The cut surface was solid, and thin bands of connective tissue

separated islands of yellow-tinged, gray-white tumor tissue which resembled the adrenal gland. The tumor was composed of large cells with a foamy cytoplasm, which resembled those of the adrenal cortex. There was no evidence of virilism of the bird, and both adrenals were normal.

*Metastasis.* Carcinomas, being malignant neoplasms, have the tendency to infiltrate or invade the surrounding tissues, and if the circumstances are propitious, new foci of growth and destruction are likely to be set up in the immediate vicinity of the parent growth or in tissues distance from the primary lesion.

In carcinoma of mammals the usual route of metastasis is by way of the lymphatics, with the blood channels involved infrequently. In chickens the lymphatic route of metastasis is apparently little utilized. As a matter of fact, true metastasis of carcinoma in chickens as a consequence of the conveyance of tumor cells from one situation to another by way of vascular channels seldom occurs. Ovarian and oviductal carcinomas early become disseminated throughout the abdominal tissues, but this results as a consequence of implantation rather than by true vascular metastasis. Distant metastatic growths may be etsablished from these tumors but are of exceptional occurrence (Fig. 30.19). A few instances have been observed in which carcinoma primary in the intestines has metastasized to the liver and to the lungs.

*Diagnostic characteristics.* Of primary importance in distinguishing carcinoma is the character of the tissue in which the tumor occurs. Being composed of epithelial elements, carcinoma should be thought of when neoplasia occurs in such tissues as the skin or the mucous membrane or in organs in which epithelial tissues are present. These tumors usually exhibit invasive or infiltrative tendencies and frequently give rise by implantation, or less often by metastasis, to secondary foci. Microscopically, although the picture is subject to marked variation, carcinoma usually is characterized by the presence of immature epithelial cells growing in an atypical infiltrative manner without the apparent guiding influence that governs normal growth or repair.

## MESOTHELIOMA

Neoplasia of the mesothelial cells covering the serous surfaces of the peritoneal cavity appears to be rare in the chicken. Olson and Bullis (1942) described one case of a nine-month-old pullet. In this case two pedunculated masses of tumor having a combined weight of 49 gm. were attached by stalks to the wall of the abdomen and the ovary. There was marked ascites. Eber and Malke (1932) mentioned four cases of what were probably mesotheliomas of the peritoneum.[15] In these cases there were numerous tumorous

---

[15] Eber and Malke designated these tumors as "endothelioma."

nodules spread over the serosa of the stomach, intestine, oviduct, and mesentery. In all cases microscopic examination revealed masses of neoplastic tissue composed of large cells which had a distinct fibroblastic character. These cells tended to form cylindrical strands, although there were regions in which the cells were loosely arranged.

Mesothelioma may arise from the serosa of the peritoneum or the pericardium. The structure of such tumors may be variable since the mesothelium is of mesodermal origin, and immature mesothelial cells may have the ability to develop into connective tissue cells. In the case of mesothelioma
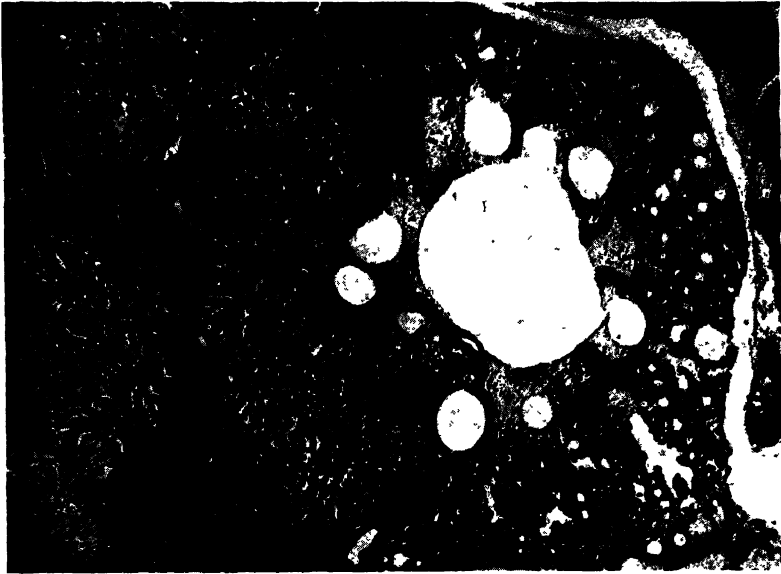


Fig. 30.19. Metastatic cystadenocarcinoma of the lungs. The neoplastic process was primary in the ovary. Same case as Figure 30.18. ×60.

described by Olson and Bullis, the polyhedral cells tended to form sheetlike masses, although in some regions they appeared to be compressed to a spindle shape. The cases described by Eber and Malke as endothelioma might be considered as mesothelioma in which the elements displayed more of the fibroblastic character than the tendency to form sheetlike masses.

Although mesothelioma appears to be a rare tumor in the chicken, it is important since it might readily become confused grossly with lymphocytoma or carcinoma. Careful study may be required to enable one to distinguish between these conditions.

### MIXED TUMORS

For convenience we have classified certain tumors of varying degrees of complexity as mixed tumors. The less complex are those whose cells,

although derived from more than one germinal layer, are in a rather advanced state of development and whose potential differentiation is subject to definite restrictions. The tumors of chickens in this category include thymoma and the so-called carcinosarcoma. A more complex group of mixed tumors is that known collectively as teratomas. These are also composed of cells derived from more than one germinal layer but differ from the less complex mixed varieties just mentioned in that the cells giving rise to a teratomatous growth have multipotent potentialities and are capable of unrestricted differentiation. In teratomatous tumors, therefore, one may recognize tissues representing all three germinal layers, some of which may have attained the specialized differentiation characteristic of adult tissues or organs. In chickens the most frequently encountered teratomatous tumors are those of the ovary, the kidney, and the testicle. A third group of complex tumors are those designated embryonal nephroma, which arise from primitive multipotent mesodermal cells that have divergent capacities for differentiation.

**Thymoma.** In chickens tumors that arise from the parenchymal tissues of the thymus are among the rarer forms of neoplasia. Feldman (1936) mentioned a case reported by Saupe and described one case of his own. In the latter instance the tumor occurred in a rooster of the Barred Plymouth Rock breed, aged two to three years, that had been killed for food. The tumor occupied the ventral aspect of the cervical region, extending from the thoracic aperture forward for a distance of 10 cm. The mass measured 10 by 5 by 4 cm. and weighed approximately 250 gm. Microscopically, the tumor consisted of a richly cellular structure composed largely of intertwining sheets or bundles of polymorphic cells. A moderate number of cells were undergoing mitotic division, and in one region concentric whorls of closely packed cells resembling Hassall's corpuscles were seen. A minimal amount of lymphoid tissue was present. The structure was well supplied with blood vascular channels. Metastasis had not occurred. It was thought that the tumor arose from the reticulum cells of the thymus. Two cases of thymoma similar to the foregoing were found by Olson and Bullis (1942).

Until additional cases provide material for the study of thymic tumors in chickens, their morphologic variations and their effects on the host will remain obscure.

**Carcinosarcoma.** A tumor somewhat similar to that which we have designated as carcinosarcoma has been described by Jackson (1936a, 1936b) as "carcinoma leiomyotosum." Jackson's term indicates an adenocarcinoma with a stroma of smooth muscle fibers. Jackson expressed the belief that the stroma of muscle fibers is merely a reaction on the part of the host to the growth of the neoplastic epithelial elements. Glietenberg (1927) described a similar process in the oviduct as a "myocystadenoma," implying that both

muscular and epithelial elements were neoplastic. Joest and Ernesti (1915) took a similar stand with respect to two tumors which they diagnosed as "adenomyoma." A third mixed neoplasm with many peritoneal implants was diagnosed by them as "sarcoma carcinomatodes." Olson and Bullis (1942) found seven cases of tumors which they termed "carcinosarcoma," all of which were widely implanted on the serosal covering of the viscera (Fig. 30.20). The epithelial elements in these cases appeared to have come from the ovary, pancreas, and adrenal. The supporting stromal tissue was considerable in amount and if seen alone would have been considered as
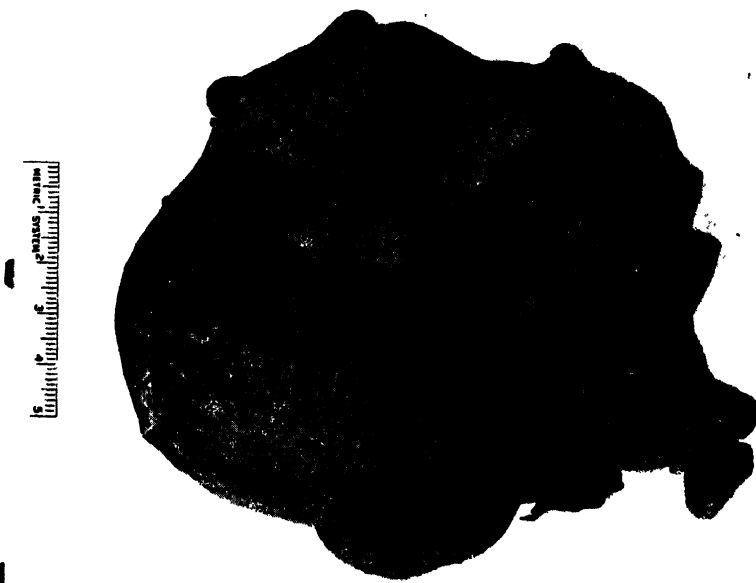


FIG. 30.20. Carcinosarcoma showing the extensive, diffuse implantation of the neoplastic tissue over the surface of the mesentery.

fibrosarcoma (Fig. 30.21). The presence of acini of epithelial cells constitutes a disquieting feature to an otherwise simple diagnosis in such cases. Bundles of smooth muscle cells also can be demonstrated in the stroma of these tumors in regions remote from where they would normally be expected. An interesting finding was a concomitant smooth muscle tumor of the mesosalpinx in four of the seven cases of carcinosarcoma. However, these leiomyomas were regarded as incidental findings.

Foulds (1937) described "mixed" tumors in the course of transplantation experiments with a carcinoma of the chicken. The original tumor was believed to have originated from the shell secreting glands of the oviduct. In one series of transplants, bone tissue was found mixed with the carcinoma

cells. Foulds discussed this feature at some length and concluded that the bone was formed either in the hyaline matrix secreted by the carcinoma cells or in the fibroblastic stroma produced by the host.

The tumors we classify as carcinosarcoma can be interpreted in different ways. They may be simple adenocarcinomas in which a highly active stromal reaction occurs and, therefore, may be considered as scirrhous adenocarcinoma. They may have originally been simple carcinomas which stimulated a stroma to the extent that it became neoplastic. On the other hand, they may have begun initially as true mixed tumors. The presence of smooth muscle tissue mixed with fibroblasts and the apparent ability of the connective tissue elements to form peritoneal implants free of the epithelial component, as well as the structure, suggest the malignant nature of the stroma. Jackson commented that such implants can be explained on the basis of assuming a primary implant of the carcinoma which became destroyed by the more rapidly growing stroma. Carcinosarcoma should not be confused with the process occasionally noted in which a connective tissue tumor infiltrates a glandular structure and causes metaplasia of the epithelial elements. Such a process might simulate the appearance of a carcinosarcoma.



FIG. 30.21. Carcinosarcoma. Tissue from an implant on the serosa of the mesentery. The essential features are the irregular acini of epithelial cells and the abundant stroma of connective tissue elements. The latter had the appearance of a malignant process. ×85.

Although there is some confusion surrounding carcinosarcomas at this time, such neoplasms as described do seem to exist as an entity in the chicken and can be distinguished from other neoplasms. Future study should clarify their position as well as determine their histogenesis.

**Teratomatous tumors.** Our experience agrees with that of Hoogland (1929) that in the chicken, teratoma and teratoid tumors occur most frequently in the ovary and in the testicle.[16] Grossly, those of the ovary have resembled carcinoma, and a microscopic examination has been necessary to reveal the true character of the neoplastic process. Usually the ovary has been involved extensively, and secondary foci representing implantation metastatic growths were commonly present. The propensity for ovarian teratoma to produce cysts containing gelatinous or mucoid material was well illustrated

---

[16] A teratoma of the ovary of a goose was recorded by Babic (1931).

in several of our cases. In two of the six cases in our collection, metastatic foci had been established in the lungs.

The microscopic appearance of these tumors is of some interest. A brief summary of the histologic characteristics of the six cases mentioned follows:

*Case 1.* The growth consisted of a mixture of (1) squamous epithelium; (2) columnar epithelium, with and without mucin; and (3) primitive mesodermal cells, with definite sarcomatous changes. Metastasis to the lung, largely of columnar epithelium, had occurred.

*Case 2.* Much of the tissue had an embryonic appearance. The tumor consisted of (1) large mucus-forming epithelial cells, which were inclined to form cysts containing mucus; (2) squamous epithelium with central areas of keratin; and (3) large amounts of sarcomatous tissue of the histiocytic variety in which mitosis was common. The pulmonary metastasis was predominantly sarcomatous.

*Case 3.* The mass was quite cystic, and the following varieties of epithelium were discernible: (1) squamous, (2) high columnar, (3) low columnar, and (4) epithelium composed of mucus-containing cells.

*Case 4.* The tumor was epithelial in character and contained squamous, tall columnar and low columnar cells.

*Case 5.* This tumor was a mixture of epithelial and undifferentiated sarcomatous elements. Epithelial cells with and without mucus and with cilia were demonstrated.

*Case 6.* There were present nests of squamous epithelial cells and regions of papillary adenocarcinoma.

In the six cases mentioned the occurrence of squamous epithelium with the formation of central regions of keratin was the most characteristic feature (Fig. 30.22). The mesodermal elements frequently exhibited a bizarre picture, with undifferentiation a notable feature. In the ovary as well as in the testicle, cells occur which have the inherent possibility of producing tissues representing all three embryonic layers. From such elements complex, jumbled, tumor-like conditions may arise that may represent an attempt to produce an embryo. However, in our material none of the ovarian teratomas contained tissue representing the entoderm; all were didermic in character. Apparently in the chicken tridermic teratomas are of rare occurrence.

We have observed one didermic teratoma of the testicle, and Jackson (1936a) mentioned a tridermic teratoma of the testicle of a rooster. Our specimen was apparently similar in many respects to that described by Olson and Bullis except that we did not find any cartilage in ours. Microscopically, the specimen we encountered showed large irregular regions of mesodermal cells in various stages of differentiation from the most primitive to well-formed histiocytic and fibroblastic types. The continuity of the mesodermal tissue was broken in many places by clefts or cysts lined with columnar epi-

thelial cells, many containing mucin. A few irregular islands of squamous epithelium were also discernible in the midst of mesodermal elements. Babic described two cases of teratoma of the testis in chickens.

The teratomas of the testicle may attain considerable size. The specimen described by Olson and Bullis measured 13 by 10 by 7 cm. and weighed 606 gm. (Fig. 30.23). In a teratoma of a twenty-one-month-old rooster described by Sheather (1911) that probably arose in the left testicle, the mass measured 15 by 11.5 by 7.5 cm. and weighed 500 gm. These tumors are usually well



Fig. 30.22. Didermic teratoma of the ovary of a one-year-old chicken. Note several epithelial "pearls" surrounded by sarcomatous stroma. ×120.

encapsulated and have an irregular nodulated surface. The color varies from grayish white to dark livid. Necrosis is common, and cysts of variable sizes are likely to be present.[17]

Teratoma of the testis has been produced experimentally in chickens by several workers (Michalowsky (Bagg, 1936); Bagg, 1936; and Falin and Gromzewa, 1940). This has been accomplished by repeated intratesticular injections of small quantities of zinc salts (chloride, sulfate, and nitrate). An associated seasonal factor is present in that teratomas of the testis can be produced only during early spring. This factor may be affected by administration of gonadotropic hormone. Baker (Foulds, 1934) succeeded in making two successful transplants of a spontaneous ovarian teratoma of the chicken.

**Embryonal nephroma.** This tumor, which we have classified perhaps more

[17] As mentioned previously, Pohl described what he designated adenocarcinoma of the testicle of a rooster that we believe was probably a teratoma. (See footnote on page 690.)

or less arbitrarily with the mixed tumors, usually originates in the kidney and is one of the commoner neoplasms of chickens.[18] Many terms have been used to designate this tumor, the term selected in most instances being that which best described the dominant structural features of the histologic process. Among the terms applied to this neoplasm are adenoma, cystadenoma, adenomyosarcoma, sarcocarcinoma, sarco-adenoma, rhabdomyo-adenosarcoma, adenosarcoma, and Wilms' tumor.

Since it is generally agreed that these tumors arise from primitive nephro-genic tissue, a term that indicates their origin rather than one which may be descriptive in a morphologic sense would seem desirable. For this reason we believe that these tumors, regardless of the variations of their structural pattern, should be designated embryonal nephroma.

*Histogenesis.* It seems a reasonable presumption that this tumor, which is character-ized by more or less unpredict-able structural variations, has its origin, regardless of anato-mic differences that may occur in different specimens, from a multipotent type of nephro-genic cell. Such a cell possesses all the capacity for differentia-tion inherent in an immature mesodermal cell.



FIG. 30.23. Teratoma of the testis. A small nod-ule of normal testicular tissue extends from the lower aspect of the tumor mass.

While the vast majority of embryonal nephromas occur in the substance of the kidney, these tumors may arise anteriorly or posteriorly, separate and removed from the nephric tissue (Feldman, 1930). The origin of embry-onal nephromas in extranephric situations may be explained on the basis of the failure of certain portions of the mesonephron to be utilized in the development of the permanent kidney. These remains of the wolffian body, instead of retrogressing, continue to grow but without the guiding influences characteristic of normal tissues, and a neoplastic process results.

*Frequency of occurrence.* The relative incidence of the occurrence of

[18] The condition also occurs in most of the other domesticated species of animals as well as in human beings. (See monograph by Feldman, 1932; p. 357.)

embryonal nephroma in chickens is uncertain. However, it is generally believed that it occurs more commonly in chickens than in mammals. Reliable statistical data on the incidence of embryonal nephroma are meager. Among a total of 113 chicken tumors exclusive of those designated "leukotic tumors" reported by Goss (1940a), four were considered embryonal nephromas. In Jackson's (1936a, p. 418) series of avian neoplasms, the embryonal nephromas accounted for 3.5 per cent of the total. Olson and Bullis (1942) found twenty-one (5.5 per cent) embryonal nephromas among 384 tumors.

There is no evidence that one sex is more susceptible to the development of embryonal nephroma than the other. Although the majority of these tumors are discovered in chickens during the young adult or adult stage of life, their histogenesis provides possibilities for their early development. In fact, it is likely that in many if not most instances these tumors are present and of demonstrable proportion when the chick is hatched. Whether or not breed is of significance in the occurrence of embryonal nephroma is problematic. Mathews (1929a) expressed the opinion that the Barred Plymouth Rock breed shows a particular predisposition for the development of this tumor.

*Sites of occurrence.* Most embryonal nephromas are found embedded in or arising from the substance of the kidney, although occasionally a specimen is encountered that has no direct contact with this organ. The tumors are usually unilateral. In thirty-one of a series of forty-one cases in our collection, one kidney only was affected, and in ten cases both kidneys were affected. Although there has been an impression that the left kidney is involved most often, in our material of thirty-one unilateral embryonal nephromas, fifteen occurred in the right kidney, fourteen occurred in the left kidney and in two instances the specific kidney affected is not known. Jackson (1936a, p. 350), however, mentioned that in four specimens in his collection in which the site of occurrence was known, three arose from the left kidney and one from the right kidney.

*Effects on the host.* As with many other neoplasms of chickens, it is difficult to determine what the effect of the presence of an embryonal nephroma may be on the well-being of the animal. Since in the chicken embryonal nephroma seldom if ever gives rise to recognizable objective symptoms, one must conclude that if only one kidney is involved and if metastasis has not occurred, the presence of a tumor of this kind is not likely to be a serious physiologic handicap. Exception must of course be taken in those instances in which the tumor is unusually large. Specimens of such size as to occupy a considerable portion of the abdominal cavity must provide a source of discomfort to the host. Paralysis may result if the tumor causes pressure on the nerves to the leg. If large hemorrhagic cysts should rupture or if there are extensive regions of necrosis, secondary disturbances may develop. If metastasis should occur, the usual effect of malignant lesions may be expected.

If both kidneys are affected and the neoplastic process is extensive and progressive, it is conceivable that sufficient nephric tissue may be destroyed to prevent adequate secretion of urine.

*Gross and microscopic description.* The gross appearance of embryonal nephroma is subject to considerable variation depending on the size and anatomic situation. The size varies within wide limits from a small focus of grayish-pink tissue embedded in the substance of the kidney to huge lobulated masses that have replaced nearly all of the nephric tissue. One of our specimens had an extranephric origin, weighed approximately 200 gm., and



FIG. 30.24. Keratinizing embryonal nephroma of the left kidney. Conglomerate masses of hard "pearly" nodules may be seen.

measured 7 by 6 by 6 cm.[19] The color may be yellow, grayish white, or grayish pink, and while most specimens have a firm fleshlike consistency, small to large cysts and foci of necrosis may provide regions that are soft and spongelike. Small, hard, pearly nodules may be recognized in some specimens (Fig. 30.24). Such dense material was found in abundance in a large keratinizing embryonal nephroma which we described some years ago (Feldman and Olson, 1933).

Although the contour of these tumors is usually irregular and frequently interrupted by fissures of variable depths, the surface of the nonkeratinizing varieties is smooth and glistening. The large keratinizing specimen previously mentioned was quite roughened, owing to the presence of innumerable closely packed, grayish-yellow, kernel-like units of variable sizes. These gave the surface of the tumors a strikingly pebbled or bosselated appearance.

[19] An exceptionally large keratinizing embryonal nephroma in a five-month-old Barred Rock cockerel was observed at the Ontario Veterinary College (1945). Two tumors were present—one large and one small. The size of the larger specimen was noteworthy. It measured 15 cm. x 10.5 cm. x 10 cm. and weighed 802 grams.

The tumors are usually well supplied with blood vessels, and in large specimens diffuse regions of hemorrhage may be present. When the interior of moderately large to large specimens is exposed by incision, the distinct lobulations are often apparent. Cysts, if present, may contain a semiclear or hemorrhagic fluid.

The multipotent embryonic character of the elements which give rise to embryonal nephromas provides for the widest possible variation of their microscopic appearance. The structure may be relatively simple or confusingly complex even within the substance of the same tumor. The most consistent feature is the great variability of their histologic appearance. No two are alike. It is difficult, therefore, to compose a written description of the microscopic features of a "typical" embryonal nephroma. In cases in which the disease is represented by the occurrence of a tiny focus just visible grossly, the essential morphologic features consist of a compact, more or less diffuse region of undifferentiated spherical or oval cells situated in the cortical zone of the kidney. Encapsulation is not evident, and the neoplastic cells at the periphery of the lesion are infiltrating into the surrounding substance of the kidney. In large specimens in which it may be presumed that the process has existed for a long period, the microscopic picture is more bizarre. The parenchyma of the tumor is highly cellular, and irregular septa of connective tissue dividing the tumor into indistinct lobules may or may not occur. The parenchyma of the process usually presents tubules or ductlike structures (Fig. 30.25). Frequently, solid nests of cuboidal cells arranged in an acinar fashion may predominate. The ductlike or tubule-like spaces are lined by single or several layers of cuboidal or columnar cells which apparently arise from the surrounding undifferentiated elements.

In some regions the undifferentiated elements, which may be cuboidal or spindle-shaped, constitute the predominant feature of the histologic picture. In other regions epithelial elements as represented by the adenomatous character of the structure are the most striking feature. Since the structure of the majority of these tumors represents a mixture of undifferentiated and differentiated cells, no useful purpose is served by attempting to determine in a given specimen which of these elements predominates.

Although the tubular spaces in many of these tumors when examined microscopically are apparently empty, in some a mucin-like substance that is probably the product of the cells lining the tubules occurs. If this substance is produced in excessive amounts, large cysts are formed. Striking evidence of the multipotent potentialities of the cells which give rise to embryonal nephroma may be found in specimens that contain in addition to the undifferentiated sarcomatous elements typical hyaline cartilage and bone. We have not recognized striated muscle in our material, although this tissue could conceivably occur.

Fairly frequently, embryonal nephromas of the chicken are encountered in which epithelial elements reveal a marked epidermoid differentiation. This occurred in varying degrees in six of twenty cases we have studied. These changes may consist of small foci of rather typical squamous cells resembling in every respect epithelial "pearls." An occasional embryonal nephroma may occur in which keratinzation is a predominant feature.

The vascular supply of the tumors is usually considerable, and hemorrhagic extravasations with organizing or organized blood clots may be present. Edema is present sometimes among the stromal elements, and a
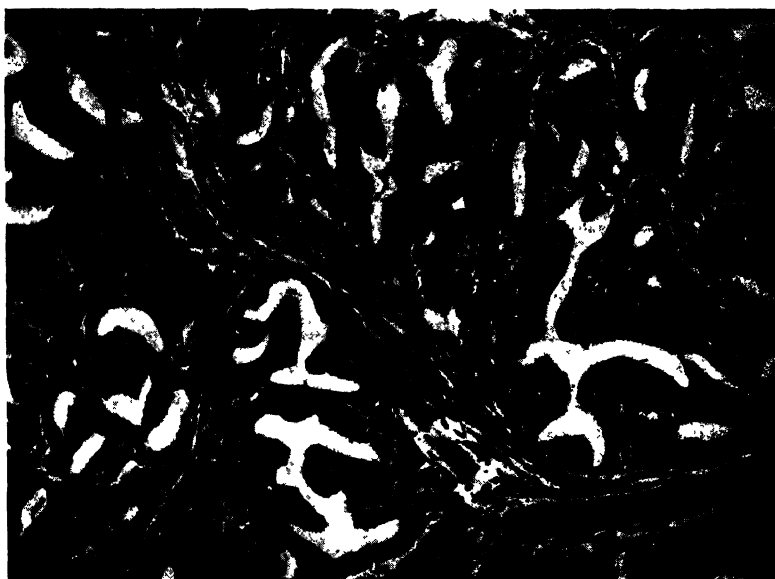


FIG. 30.25. Embryonal nephroma, kidney of a chicken. Epithelial elements predominate. ×150.

few to large numbers of eosinophilic granulocytes may occur promiscuously throughout the tumor.

*Metastasis.* Although mitosis and other evidences of instability and progression are commonly observed in embryonal nephroma, metastatic dissemination of these tumors appears to be relatively infrequent. Metastasis has been recorded by Jackson (1936a) and by Mathews (1929) and by Duran-Reynals (1946b), but has not been demonstrated in the material at our disposal. In our experience most of the larger specimens have been rather well encapsulated, and while infiltration of the neoplastic elements into the substance of the kidney occasionally occurs, this is infrequent. As a matter of fact, the fibrous zone of connective tissue that is usually present between the neoplastic and the nephric elements must be considered a

formidable barrier against the invasive tendencies of the tumor. The destructive effect on the kidney of many embryonal nephromas is due to the pressure atrophy and other retrograde influences that ensue as a consequence of the gradual, progressive expansion of the growth. It should be kept in mind, however, that these are at least locally malignant tumors, and most if not all of these should be capable of setting up metastatic foci if circumstances are propitious.

*Transplantability.* Duran-Reynals (1946b) attempted transplants from ten spontaneous cases, seven of which proved nontransplantable. One yielded a fibroma type of growth, and grafts from another resembled the original embryonal nephroma with sarcomatous features. Neither of these could be carried in serial passage. The third transplantable embryonal nephroma was carried in regular serial passage as a sarcoma. Duran-Reynals and Shrigley (1946) are inclined to regard this as an independent sarcoma since a cell-free etiological agent was demonstrated.

*Diagnostic characteristics.* In chickens, as Jackson (1936a, p. 351) pointed out, embryonal nephroma should always be considered when a tumor occurs in or near the kidney or when a tumor is encountered, suspended from the lumbar region although somewhat removed from the kidney. Microscopically, these tumors are usually entirely unlike any other neoplasms. The mixture of undifferentiated cuboidal or spindle cells and of adenomatous epithelial structures, keratin, cartilage, or bone should be sufficient for their correct identification.

### CONCOMITANT NEOPLASIA

Concomitant or multiple neoplasia, in which two or more distinct types of neoplastic disease occur in the same bird, appears to have received little attention in the past. Atlhough multiple neoplasia has been noted in other animals, especially old dogs (Feldman, 1932), only a few comments were found in the literature on concomitant neoplasia of the chicken. Perhaps the fact that relatively few chickens are allowed to attain advanced age may be a factor in the apparent infrequency of multiple neoplasia. Another explanation for the relatively few observed cases is that they are not recognized since relatively few diagnoses of spontaneous avian neoplasia are made with the aid of histologic examination. Thorough histologic examination is obviously necessary for the recognition of instances of concomitant neoplasia, for without such an examination, unexamined foci of tumor may be assumed to represent metastasis of a primary growth.

Jackson (1936a) discussed "collision" tumors in which elements of two distinctly different neoplastic processes become intermingled to create the impression of a "mixed" tumor. He described an instance of a histiocytic sarcoma and myelocytoma occurring in the same chicken. Babic (1931) de-

scribed a chicken which had a fibroma of the intestine in addition to hemangioma of the liver, kidney, and serosa of the proventriculus. Jármai (1939) found both fowl leukosis and fibrosarcoma as concomitant tumors in a parakeet.

Olson and Bullis (1942) observed nineteen instances of concomitant neoplasia in a collection of 384 neoplasms found in 365 chickens. Of six chickens that had lymphocytoma, four also had embryonal nephroma, one had neurogenic sarcoma, and one had leukosis. Of three chickens that had fowl leukosis, two also had myelocytoma, and one had fibrosarcoma. Of six birds that had leiomyoma, four were also affected with carcinosarcoma, and two with carcinoma. Myelocytoma and embryonal nephroma were found in one chicken. Histiocytic sarcoma and hemangioma were concomitant tumors in one chicken; adenoma of the thyroid and melanoma of the tongue, in another; and a third chicken had an adenoma of the pancreas and a fibrosarcoma in the pelvic cavity.

The possibility of an etiologic relation between neoplasms occurring simultaneously in the same chicken is suggested in cases of concomitant neoplasia. However, with the possible exception of fowl leukosis and fibrosarcoma, there is little evidence to support such an interpretation. Experimentally, several strains of the agent responsible for fowl leukosis have been shown to be capable of inducing fibrosarcoma. The spontaneous occurrence of this combination of concomitant neoplasia, therefore, may not be entirely due to chance.

Since certain types of spontaneous tumors are relatively common in the chicken, the occurrence of the more common varieties in birds of a group under experimental observation may be anticipated. This probability necessitates extensive and careful provision for the control of experiments designed to attempt transmission of neoplasia in chickens. Concomitant neoplasia may occur in such experimental birds. Usually such cases should be interpreted as a spontaneous neoplasm which occurred in a bird in which a second type of neoplasia also had developed as the result of experimental inoculation. An example is the association of lymphocytoma and fowl leukosis in a chicken which had received material containing the causative agent of fowl leukosis. Olson (1936) observed two such instances and regarded the lymphocytoma as of spontaneous origin and unrelated to the fowl leukosis.

Concomitant neoplasia may develop in experimental birds treated with carcinogenic agents. Murphy and Sturm (1941a) reported the finding of leukemia and adenocarcinoma in a chicken that had received intramuscular injections of dibenzanthracene in lard. The dibenzanthracene was regarded as responsible for the development of both neoplastic processes.

## TUMORS OF BIRDS OTHER THAN CHICKENS

Various types of neoplastic disease have been reported from different species of both wild and domesticated birds. Although adequate data are not available to make absolute comparisons, in no other single species of bird do tumors appear as likely to develop as in the domestic chicken.

The observations at the University of Leipzig from 1899 to 1931 indicate the relative incidence of neoplasia among various domesticated species of birds (Eber and Malke, 1932). During this period 2,353 pigeons were submitted to necropsy, and fourteen were affected with neoplastic disease, an incidence of 0.6 per cent. Seven hundred and twenty geese were examined, one of which had "chondrosarcoma myxomatodes." Among 692 ducks, one had adenocarcinoma of the liver; among 459 turkeys examined, two had neoplasms of the liver (mixed cell type of sarcoma) ; and among 204 pheasants, one had adenoma of the lung, and one had myxosarcoma of the liver. In the same interval of time fifty-three guinea fowl, fifty-two peacocks, and twenty-five swans were examined, in none of which was neoplastic disease found. A total of 11,903 chickens were examined, and 3.12 per cent (371) had tumors. Babic (1931) in Yugoslavia, reported sixteen cases of neoplasia in birds other than chickens. These were encountered during the period between 1923 and 1931. In this interval fifty-nine pigeons were examined, five of which had tumors; twenty-seven parakeets, three of which had tumors; forty-five canaries, one of which had tumor; forty geese, two of which had tumors; twenty-nine turkeys, two of which had tumors; ten pheasants, one of which had a tumor; fourteen owls, of which one had a tumor; two storks, of which one had a tumor. Of eighty-one ducks examined, none revealed the presence of neoplasia.

Fox (1923) listed forty-four neoplasms found in captive wild birds. Eleven varieties of tumor were identified. Species most commonly affected were in the family of Psittacidae and accounted for twenty-three of the cases, sixteen of which were in the undulated grass parakeet (*Melopsittacus undulatus*). It is of interest to note that in a bird of this species Jármai (1939) found a sarcoma and fowl leukosis. Epidermoid carcinomas of the feet of wild birds were reported by Emmel (1930).

Neoplasms in other species of birds are quite similar in general character to those found in chickens. Most reports in the literature concern cases of epithelioblastoma and tumors of connective tissue. Neoplasia of the lymphoid cell system may be encountered occasionally in various species of birds, especially turkeys, but in none is it as common as in the chicken.

## REFERENCES

Abels, H.: 1929. Die Geschwülste der Vogelhaut. Zeitschr. f. Krebsforsch. 29:183.

Apperly, F. L.: 1935. Primary carcinoma of the lung in the domestic fowl. Am. Jour. Cancer 23:556.

Ask-Upmark, E.: 1938. Ein gehäuftes ("epidemisches") Vorkommen von Hühnertumoren. Frankfurt. Zeitschr. f. Path. 52:51.

Babic, I.: 1931. Spontani tumori peradi. Veterinarski Archiv. 1:158.

Bagg, H. J.: 1936. Experimental production of teratoma testis in the fowl. Am. Jour. Cancer 26:69.

Baker, S. L.: Quoted by Foulds.

Ball, R. F.: 1945. Two unusual neoplasms in the chicken iris. Cornell Vet. 35:383.

Berner, O.: 1923. Virilisme surrénal chez une poule. Rev. franc. d'endocrinol. 1:474.

Boyd, W.: 1938. A Text-book of Pathology; An Introduction to Medicine. Lea and Febiger, Philadelphia, pp. 42–45; 320–25.

Brewer, N. R., and Brownstein, B.: 1946. The transmission of lymphomatosis in the fowl. Am. Jour. Vet. Res. 7:123.

Burmester, B. R.: 1945. The incidence of lymphomatosis among male and female chickens. Poultry Sci. 24:469.

——— and Belding, T. C.: 1947. Immunity and cross immunity reactions obtained with several avian lymphoid tumor strains. Am. Jour. Vet. Res. 8:128.

——— and Nelson, N. M.: 1945. The effect of castration and sex hormones upon the incidence of lymphomatosis in chickens. Poultry Sci. 24:509.

——— and Prickett, C. O.: 1945. The development of highly malignant tumor strains from naturally occurring avian lymphomatosis. Cancer Res. 5:652.

Campbell, J. G.: 1943. Histocytic and fibroplastic sarcoma (mixed-cell sarcoma) in the domestic fowl. Jour. Comp. Path. and Therap. 53:323.

———: 1945. Neoplastic disease of the fowl with special reference to its history, incidence, and seasonal variation. Jour. Comp. Path. and Therap. 55:308.

Carr, J. G.: 1943. Prolonged antibody production following recovery of fowls from Rous No. 1 sarcoma. Brit. Jour. Exper. Path. 24:138.

———: 1944. Lack of transmission of avian tumor virus from carrier hens to their offspring via the egg. Proc. Roy. Soc. Edinburgh, Sec. B, 62:54.

Claude, A., and Murphy, J. B.: 1933. Transmissible tumors of the fowl. Physiol. Rev. 13:246.

Cole, R. K.: 1946. An avian retinoblastoma. Cornell Vet. 36:350.

Curtis, M. R.: 1915. Frequency of occurence of tumors in the domestic fowl. Jour. Agr. Res. 5:397.

Davis, O. S., and Doyle, L. P.: 1947a. Studies in avian leucosis. I. The transmissibility of visceral lymphomatosis. Am. Jour. Vet. Res. 8:103.

——— and Doyle, L. P.: 1947b. Studies in avian leucosis. II. The use of biopsy technique in the study of visceral lymphomatosis. Am. Jour. Vet. Res. 8:113.

Duran-Reynals, F.: 1940. Neutralization of tumor viruses by the blood of normal fowls of different ages. Yale Jour. Biol. Med. 13:61.

———: 1946a. Transplantability and presence of virus in spontaneous sarcomas and fibromas of chickens in relation to the age of the tumor-bearing animal. Cancer Res. 6:529.

———: 1946b. On the transplantability of lymphoid tumors, embryonal nephromas, and carcinomas of chickens. Cancer Res. 6:545.

——— and Shrigley, E. W.: 1946. A study of five transplantable chicken sarcomas induced by viruses. Cancer Res. 6:535.

Eber, A., and Kriegbaum, A.: 1916. Untersuchungen über Eierstocks- und Eileitergeschwülste beim Haushuhn. Zeitschr. f. Krebsforsch. 15:404.

——— and Malke, E.: 1932. Geschwulstbildungen beim Hausgeflügel. Zeitschr. f. Krebsforsch. 36:178.

Ehrenreich, M., and Michaelis, L.: 1906. Über Tumoren bei Hühnern. Zeitschr. f. Krebsforsch. 4:586.

Ellermann, V.: 1920. Histogenese der übertragbaren Hühnerleukose. I. Die myeloische Leukose. Folia haemat. 26:135.

——— and Bang, O.: 1908. Experimentelle Leukämie bei Hühnern. Zentralbl. f. Bakt. I. Orig. 46:595.

Elsner: Quoted by Reis and Nobrega.

Emmel, M. W.: 1930. Epidermoid cancers on the feet of wild birds. Jour. Am. Vet. Med. Assn. 77:641.

Engelbreth-Holm, J.: 1942. Spontaneous and Experimental Leukaemia in Animals. Oliver and Boyd, London, 245 pp.

——— and Rothe Meyer, A.: 1932. I. Bericht über neue Erfahrungen mit einem Stamm Hühner-Erythroleukose. Acta Path. et microbiol. Scandinav. 9:293.

Engelbreth-Holm, J., and Rothe Meyer, A.: 1935. On the connection between erythroblastosis (haemocytoblastosis), myelosis, and sarcoma in chicken. Acta Path. et microbiol. Scandinav. 12:352.

Ewing, J.: 1928. Neoplastic Diseases; A Treatise on Tumors. 3rd Ed. W. B. Saunders and Co., Philadelphia, Chap. 16, pp. 240–56.

Falin, L. I., and Gromzewa, K. E.: 1940. Zur Pathogenese der experimentellen teratoiden Geschwülste der Geschlechtsdrüsen. II. Mitteilung. Teratoide Geschwülste der Geschlechtsdrüsen bei Hähnen, erzeugt durch Injektionen von Zn (NO₃)₂ Lösung. Virchows Arch. f. path. Anat. 306:578. Abstr. Cancer Res. 1:580. (1941.)

Feldman, W. H.: 1930. Extranephric embryonal nephroma in a hog. Jour. Cancer Res. 14:116.

———: 1932. Neoplasms of Domesticated Animals. W. B. Saunders Co., Philadelphia, pp. 51–53; 178–93; 357.

———: 1936. Thymoma in a chicken (Gallus domesticus). Am. Jour. Cancer 26:576.

——— and Olson, Jr., C.: 1933. Keratinizing embryonal nephroma of the kidneys of the chicken. Am. Jour. Cancer 19:47.

Fischel, Alfred: 1922. Zur Eröffnung des neuen Institutes fur Embryologie. Wien. klin. Wochenschr. 35:355.

Foulds, L.: 1934. The filtrable tumours of fowls. A critical review. Suppl. to 11th Scientific report of Cancer Res. Fund, London, 41 pp.

———: 1937. A transplantable carcinoma of a domestic fowl, with a discussion of the histogenesis of mixed tumors. Jour. Path. and Bact. 44:1.

Fox, H.: 1923. Disease in Captive Wild Mammals and Birds. J. B. Lippincott Co., Philadelphia, pp. 462–82.

Friedgood, H. B., and Uotila, U. U.: 1941. Occurrence of ovarian "tumors" in spontaneous virilism of the hen. Endocrinology 29:47.

Furth, J.: 1931. Erythroleukosis and the anemias of the fowl. Arch. Path. 12:1.

———: 1933. Lymphomatosis, myelomatosis, and endothelioma of chickens caused by a filtrable agent. I. Transmission experiments. Jour. Exper. Med. 58:253.

———: 1935. Lymphomatosis in relation to fowl paralysis. Arch. Path. 20:379.

Glietenberg: 1927. Ein Myokystadenom des Eileiters eines Huhnes. Zeitschr. f. Fleisch. u. Milchhyg. 37:135.

Goldberg, S. A.: 1919. The differential features between melanosis and melanosarcoma. Jour. Am. Vet. Med. Assn. 56:140–53; 250–64.

Goss, L. J.: 1940a. The incidence and classification of avian tumors. Rep. N. Y. St. Vet. Coll. (1939–40): 103.

———: 1940b. The incidence and classification of avian tumors. Cornell Vet. 30:75.

Hamilton, C. M., and Sawyer, C. E.: 1939. Transmission of erythroleukosis in young chickens. Poultry Sci. 18:388.

Heim, F.: 1931. Hühnergeschwülste. Zeitschr. f. Krebsforsch. 33:76.

Hoogland, H. J. M.: 1929. In van Heelsbergen, T., Handbuch der Geflügelkrankheiten und der Geflügelzucht. Ferdinand Enke, Stuttgart, pp. 484–97.

Hutt, F. B., Cole, R. K., and Bruckner, J. H.: 1941. Four generations of fowls bred for resistance to neoplasms. Poultry Sci. 20:514.

Jackson, C.: 1936a. The incidence and pathology of tumours of domesticated animals in South Africa: A study of the Onderstepoort collection of neoplasms with special reference to their histopathology. Onderstepoort Jour. Vet. Sci. 6:1.

———: 1936b. Neoplastic diseases in poultry. Jour. So. African Vet. Med. Assn. 7:69.

Jármai, K.: 1934. Die Leukosen der Haustiere. Ergebn. d. allg. Path. u. path. Anat. 28:227.

———: 1935. Tumorerzeugung mit dem Leukoseagens der Hühner. Arch. f. wiss. u. prakt. Tierheilk. 69:275.

———: 1938. Über die Wirksamkeit der Eiweissfraktionen bei der übertragbaren Hühnerleukose. Arch. f. wiss. u. prakt. Tierheilk. 73:295.

———: 1939. Leukose und Sarkom beim Wellensittich. Arch. f. wiss. prakt. Tierheilk. 74:316.

Joest, E., and Ernesti, S.: Quoted by Heim.

——— and Ernesti, S.: 1915–1916. Untersuchungen über spontane Geschwülste bei Vögeln mit besonderer Berücksichtigung des Haushuhns. Zeitschr. f. Krebsforsch. 15:1.

Johnson, E. P.: 1934. The etiology and histogenesis of leucosis and lymphomatosis of fowls. Va. Agr. Exper. Sta., Tech. Bul. 56.

Jordan, H. E.: 1936. The relation of lymphoid tissue to the process of blood production in avian bone marrow. Am. Jour. Anat. 59:249.

Jungherr, E., and Wolf, A.: 1939. Gliomas in animals; a report of two astrocytomas in the common fowl. Am. Jour. Cancer 37:493.

McGowan, J. P.: 1928. On Rous, Leucotic and Allied Tumours in the Fowl: A Study in Malignancy. The Macmillan Co., New York.

Marine, D., and Rosen, S. H.: 1940. Increase in the incidence of lymphomatosis in male fowls by castration. Am. Jour. Cancer 39:315.

—— and Rosen, S. H.: 1941. Sex hormones and lymphomatosis in fowls. Proc. Soc. Exper. Biol. and Med. 47:61.

Mathews, F. P.: 1929a. Adenosarcomata of the kidneys of chickens. Jour. Am. Vet. Med. Assn. 74:238.

——: 1929b. Leukochloroma in the common fowl. Its relation to myelogenic leukemia and its analogies to chloroma in man. Arch. Path. 7:442.

—— and Walkey, F. L.: 1930. Hypernephromas in the common fowl. Jour. Am. Vet. Med. Assn. 77:218.

Meyer: Quoted by Feldman, W. H. (1932).

Michalowsky, I. O.: Quoted by Bagg, H. J.

Murphy, J. B., and Sturm, E.: 1941a. Further investigation of induced tumors in fowls. Cancer Res. 1:477.

—— and Sturm, E.: 1941b. Further investigation on transmission of induced tumors in fowls. Cancer Res. 1:609.

Nelson, N. M.: 1946. Leiomyoma of the ventral ligament of the oviduct of the chicken. Am. Jour. Path. 22:1047.

Nyfeldt, A.: 1934. Étude sur les leucoses des poules. I. Une myéloblastose pure. Sang. 8:566.

Oberling, C., and Guérin, M.: 1934a. La leucémie érythroblastique ou érythroblastose transmissible des poules. Bul. Assoc. franc. p. l'étude du cancer 23:38.

—— and Guérin, M.: 1934b. Formations de moelle osseuse hétérotopique d'aspect tumoral chez la poule. Bul. Assoc. franc. p. l'étude du cancer 23:341.

Olson, Jr., C.: 1936. A study of transmissible fowl leukosis. Jour. Am. Vet. Med. Assn. 89:681.

——· 1940. Transmissible fowl leukosis. A review of the literature. Mass. Agr. Exper. Sta., Bul. 370.

——: 1941. A transmissible lymphoid tumor of the chicken. Cancer Res. 1:384.

——: 1942. A study of neoplastic diseases in a flock of chickens. Am. Jour. Vet. Res. 3:111.

——: 1945. The immunizing action of a lymphoid tumor in chickens. Am. Jour. Vet. Res. 6:103.

——: 1947. Immunization against a lymphoid tumor of the chicken. IV. Use of miscellaneous tissues. Cornell Vet. 37:231.

—— and Bullis, K. L.: 1942. A survey and study of spontaneous neoplastic diseases in chickens. Mass. Agr. Exper. Sta., Bul. 391.

—— and Dukes, H. H.: 1938. The basal metabolic rate of chickens affected with fowl paralysis, transmissible fowl leukosis and certain spontaneous neoplasms. Folia Haemat. 60:57.

Ontario Department of Agriculture, 1945: Report of the Ontario Veterinary College Sessional, Paper No. 29, 1945. Printed by order of the Legislative Assembly of Ontario, Toronto.

Oshima, F., and Roki, K.: 1925. Studien über die Hühnergeschwülste. Trans. Jap. Path. Soc. 15:279.

Peacock, P. R.: 1946. The etiology of fowl tumors. Cancer Res. 6:311–28.

Penfield, W.: 1932. Cytology and Cellular Pathology of the Nervous System. P. B. Hoeber Inc., New York, 3:905.

Pentimalli, F.: 1915–1916. Ueber die Geschwülste bei Hühnern. I. Mitteilung. Allgemeine Morphologie der spontanen und der transplantablen Hühnergeschwülste. Zeitschr. f. Krebsforsch. 15:111.

——: 1941. Transplantable lymphosarcoma of the chicken. Cancer Res. 1:69.

Petit, G., and Germain, R.: Quoted by Pohl.

Peyron, A., and Blier, J.: 1927. Sur un nouveau cas de tumeur transplantable chez les oiseaux. Myome malin chez un coq. Bul. de l'assoc. franc. pour l'étude du cancer. 16:516.

Pohl, R.: 1926. Beiträge zur Pathologie der beim Haushuhne auftretenden Geschwülste. Arch. f. wiss. u. prakt. Tierheilk. 54:142.

Prospero: Quoted by Heim.

Reinhardt, R.: 1930. Die pathologisch-anatomischen Veränderungen bei den spontanen Krankheiten der Hausvögel. Ergebn. der allg. Path. u. path. Anat. 23:553.

Reis, F., and Nobrega, P.: 1936. Tratado de Doencas das Aves. Instituto Biologico. São Paulo, Brazil, p. 77.

Reitsma: Quoted by Hoogland.

Savage, A.: 1926. Adenocarcinoma of gallbladder in a hen. Cornell Vet. 16:67.

Schneider, M.: 1926. On the frequency of spontaneous tumors in the domestic fowl. Jour. Exper. Med. 43:433.

Schöppler, H.: 1913. Carcinoma ventriculi cylindrocellulare beim Haushuhn. Zeitschr. f. Krebsforsch. 13:332.

Seifried, O.: 1923. Das "Oophoroma folliculare" beim Huhn. Ein Beitrag zur Histogenese der epithelialen Ovarialtumoren. Zeitschr. f. Krebsforsch. 20:188.

Sheather, A. L.: 1911. Teratoma in a cock. Jour. Comp. Path. and Therap. 24:129.

Teutschlaender, O.: Quoted by Heim.

Wickware, A. B.: 1946. The incidence of erythroleucosis following inoculation by various routes. Canad. Jour. Comp. Med. 10:74.

Zannini: Quoted by Heim.

CHAPTER THIRTY-ONE

## EXTERNAL PARASITES OF POULTRY

By E. A. BENBROOK, *Department of Veterinary Pathology,
Iowa State College, Ames, Iowa*

☆   ☆   ☆

### INTRODUCTION

Ectoparasites of poultry comprise a relatively large group. Certain species are well known, but it is difficult to evaluate the importance of some because their distribution has not as yet been accurately determined. Furthermore, those parasites believed today to be of relatively minor importance may prove later to need more attention as their number increases or as they become more widely distributed or recorded.

Before considering the external parasites individually, it appears advisable to review the group. This will orient them with relation to the internal parasites and to the animal kingdom in general.

Basically, external parasites are all those living forms which, for the purpose of securing food, live on the exterior of the host's body. Thus might be included not only animal parasitic forms but also parasites belonging to the plant kingdom, such as the bacteria, molds, fungi, and yeasts, certain of which attack the skin or feathers. However, of chief concern are those *animal* forms that live as parasites on birds. Even some of these do not confine themselves entirely to the surface; although probably at some remote period of evolution they were strictly external parasites. Examples of this peculiarity include the scaly-leg mite that tunnels into the epithelium of the lower legs, certain quill mites that enter the quill bases, and the subcutaneous and air-sac mites that live beneath the skin and in the internal organs, respectively.

Certain ectoparasites of birds actually eat the dead cells of the skin and its appendages, e.g., lice. However, for most of them the skin merely serves as a convenient medium through which they draw blood or lymph and from which they obtain warmth and shelter.

Ectoparasites may be closely confined to their hosts during the entire life cycle, as is true for bird lice, transmission taking place by host contacts. Others wander freely from bird to bird. Some are highly host specific, which contradicts the viewpoint that chicken lice, for example, propagate on horses or other animals. On the other hand, some species may maintain a rather

loose relationship to their food supply. Adapted as they are to living on birds, they do not always confine their activities to one particular host species or even to birds as a group. Such forms include certain of the host-cosmopolitan forms: the gnats, mosquitoes, bedbugs, and fleas. Other external parasites, e.g., the fowl tick and the common red mite, attack birds only at night, hiding in surrounding shelters during the daytime.

Variations in habits such as noted above are important when control measures are to be considered. Mites, as a group, cannot be successfully controlled by any single method of attack because of habit variations among species. This indicates the necessity for accurate identifications as a preliminary. In case of doubt the various state and national diagnostic services may be called upon for assistance.

## CLASSIFICATION

Practically all external parasites of birds belong in the invertebrate animal group (phylum) **ARTHROPODA**. The arthropods are jointed-limbed animals without a vertebral column. Nearly all those parasitic on birds and on other animals are further characterized by having tracheal tubes for breathing.

Arthropods with antennae include the parasitic insects (class **INSECTA**) , such as lice, bedbugs, fleas, beetles, flies, and gnats. Of these, only the last three have wings. The insect body is divided into head, thorax, and abdomen. The legs and the wings (if present) are attached to the thorax. Insects are further distinguished by having three pairs of legs in the adult stage.

Arthropods not having antennae include the spider-related arachnids (class **ARACHNIDA**) of which many of the mites and all of the ticks are parasites. The arachnid body consists of a combined head and thorax (cephalothorax) not usually marked off from the unsegmented abdomen. The legs of arachnids are attached to the cephalothorax. There are typically four pairs of legs in the adult and nymphal stages, whereas the larval arachnid is provided with only three pairs.

## CONTROL IN GENERAL

Because of the comparatively low economic value of most individual poultry, it has long been hoped that simple, rapid, and inexpensive methods might be found for the control of external (and internal) parasites. For the present it must suffice to control them by persistent destruction on the birds and in the birds' surroundings, using external applications and preventive measures.

Parman (1928) and associates carefully tested thirty substances in an attempt to control external parasites of poultry through internal medication via feed or drinking water. None of these substances was of practical value.

More recently Emmel (1937) tried to control external parasites of poultry by feeding sulfur. The results on a limited number of birds appeared hopeful and indicated the need for further research.

Creighton, Dekle, and Russell (1943) found internal sulfur therapy to be ineffective for louse control, using 5 to 10 per cent sulfur in the mash. What little control occurred was the result of the birds getting some of the sulfur-feed mixture in the feathers or on the skin.

The use of artificial nest eggs containing antiparasitic substances has not been efficient, and bad effects on both the birds and their eggs were noted.

Frequent inspection of birds and their surroundings, persistent cleanliness, proper nutrition, and the use of those chemical agents that are supported by authentic research will effectively control external parasites in most instances.

## LICE OF POULTRY

These insects are the most common and widespread external parasites of birds. They all belong in the order MALLOPHAGA, those lice having chewing mouthparts. Most authorities agree that there are more than forty species of lice reported from domesticated fowls. A survey of the literature, however, shows considerable variance in the listing of North American species and a decided disagreement on the scientific names assigned to them. Fortunately, as far as the veterinarian and the poultry raiser are concerned, the various species of bird lice are at present all controlled by the same methods. Birds frequently harbor several species of lice at the same time.

The more commonly reported lice of North American poultry are as follows:

*Chicken lice:*
> *Eomenacanthus stramineus,* body louse
> *Menacanthus latus,* large body louse
> *Menopon gallinae,* shaft or small body louse
> *Lipeurus heterographus,* head louse
> *Lipeurus caponis,* wing louse
> *Lipeurus bishoppi,* wing louse
> *Lipeurus angularis,* wing louse
> *Goniocotes gigas,* large chicken louse
> *Goniocotes hologaster,* fluff louse
> *Goniodes dissimilis,* brown chicken louse

*Turkey lice:*
> *Eomenacanthus stramineus,* body louse
> *Menopon gallinae,* shaft or small body louse
> *Lipeurus gallipavonis,* slender turkey louse
> *Goniodes meleagridis,* large turkey louse

*Guinea fowl lice:*
    *Menopon gallinae,* shaft or small body louse
    *Goniocotes gigas,* large chicken louse
    *Goniocotes hologaster,* fluff louse
    *Lipeurus numidae,* guinea fowl louse

*Duck and goose lice:*
    *Anaticola crassicornis,* duck louse
    *Anatoecus dentatus,* duck and goose louse

*Pigeon lice:*
    *Columbicola columbae,* slender pigeon louse
    *Goniocotes bidentatus,* small pigeon louse
    *Goniodes damnicornis,* little feather louse
    *Colpocephalum turbinatum,* narrow body louse

Lousiness (pediculosis) of birds is diagnosed by finding on the birds wingless, dorsoventrally flattened, brownish-yellow, quickly moving insects. In size, bird lice vary from somewhat less than 1 mm. in length to the largest species which are slightly more than 6 mm. long. (Figs. 31.1, 31.2, 31.3, 31.4, and 31.5). See also line drawings in Whitehead (1942).

Lice spend the entire life cycle on the host. Eggs are attached, often in clusters, to the feathers. The entire life cycle takes from about two to three weeks for completion. One pair of lice may produce 120,000 descendents within a period of a few months. Their normal life span is several months, but away from the birds they can remain alive only 5 or 6 days.

Although bird lice ordinarily eat cast-off bits of skin and feathers and fragments of feces that adhere around the vent, it has been shown by Wilson (1933) that *Eomenacanthus stramineus,* the body louse of chickens, may puncture soft quills near the bases and consume the blood that oozes out. This was confirmed by Crutchfield and Hixson (1943), and, in addition, they stated that the body louse draws blood by gnawing through the covering layers of the skin itself.

Lousiness is characterized by unthriftiness and failure to gain normal weight. The reason for these symptoms appears to be explained by the constant irritation to the nerve endings in the skin. Birds, like other animals, especially when young, need rest to overcome physiologic fatigue. Lice prevent rest and sleep, thereby interfering with the normal metabolic processes.

Because it is so commonplace, lousiness in domesticated poultry is frequently overlooked as a factor in disease. Young birds may die when heavily infested. Birds in general may be predisposed to attack by bacterial and protozoan disease agents.
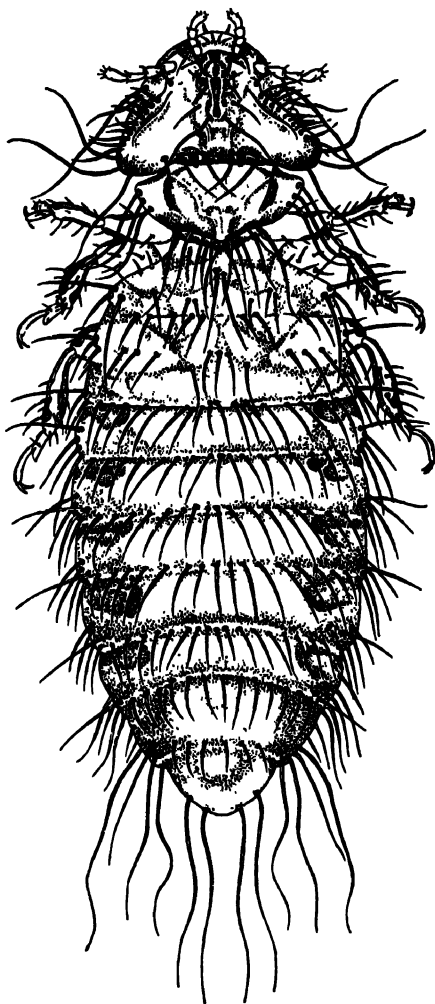
FIG. 31.1. *Menopon* sp. Chicken louse, male. ×53. (Reis and Nobrega.)
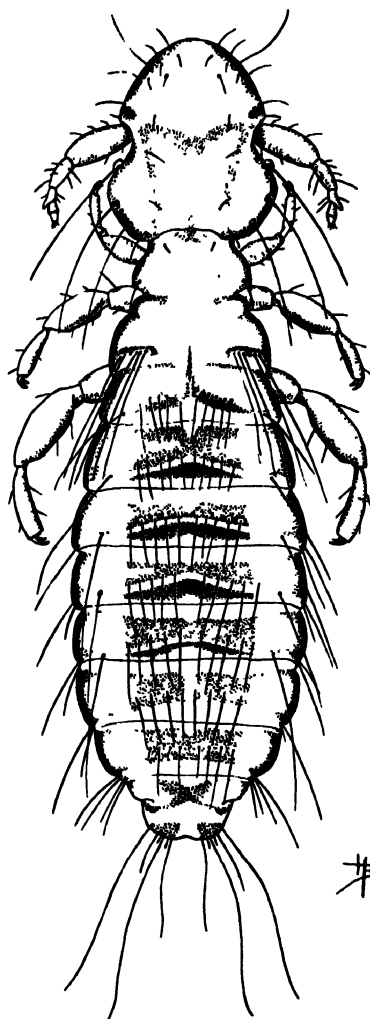
FIG. 31.2. *Lipeurus heterographus.* Head louse of chickens, male. Greatly enlarged. (U.S.D.A., Bur. Entomol. and Plant Quar.)

**Control of bird lice.** This problem primarily involves the birds themselves, although in heavy infestations the houses should be cleaned and disinfested. Lice multiply most rapidly during cold weather when birds are in closer contact; therefore, it is essential to combat them during the mild days of the fall season so as to allow birds to go into production in the best of health. Once a flock is free from lice, it is important to isolate new additions until their freedom from these parasites has been assured.

*Control methods* include (1) fumigation, (2) application of medicated

ointments, (3) wet-dipping, (4) dusting or dry-dipping. Whatever method is chosen, it should be repeated at about 10-day intervals as often as may be necessary. Many substances are available as treatments, the choice of which depends upon availability and the method of husbandry in use.

*Fumigation methods* most widely used include:

A. Fumigation with nicotine sulfate 40 per cent solution. This is painted on the roosts at the rate of about 8 ounces per 100 feet, just before the birds retire for the night. It should be used on still nights when the temperature is above 60° F. Some ventilation of the house is necessary. The heat of the birds' bodies volatilizes the nicotine which permeates the feathers. Head lice may not be reached by this method.

B. Fumigation with orthophenylphenol. One-fifth of a pound of the chemical is added to 1 gallon of kerosene. This is brushed or sprayed on the roosts as noted for nicotine sulfate solution, according to Chandler (1930).

C. Fumigation with naphthalene. Powders containing 10 to 20 per cent naphthalene may be sprinkled over the backs of roosting birds. It should not be placed in nests in any form. Naphthalene should not be used in the form of



FIG. 31.3. *Goniocotes* sp. Chicken louse, male. ×20. (Reis and Nobrega.)

moth balls. (See chapter on Poisoning.)

*Ointments for lice* include:

A. Mercurial ointment. This is effective only for young birds such as chicks and poults. It must not be used on breeding stock for two to three months prior to or during the breeding season because the mercury will affect fertility of the birds and eggs (Deakin and Robertson, 1933). Metallic mercury 5 per cent is incorporated in petrolatum and dabbed on the back of the head, around the vent and under the wings.

B. Nicotine sulfate ointment consists of one part of 40 per cent nicotine sulfate in 50 parts of petrolatum. It is used as was recommended for mercurial ointment.

*Wet-dipping methods* include:

A. Sodium fluosilicate or sodium fluoride. Either of these substances may be added to warm water at the rate of 1 ounce per gallon. One ounce of near-neutral soap per gallon of dip increases its efficiency (Telford, 1945a). Dipping should be carried out on a mild sunny day or in a warm
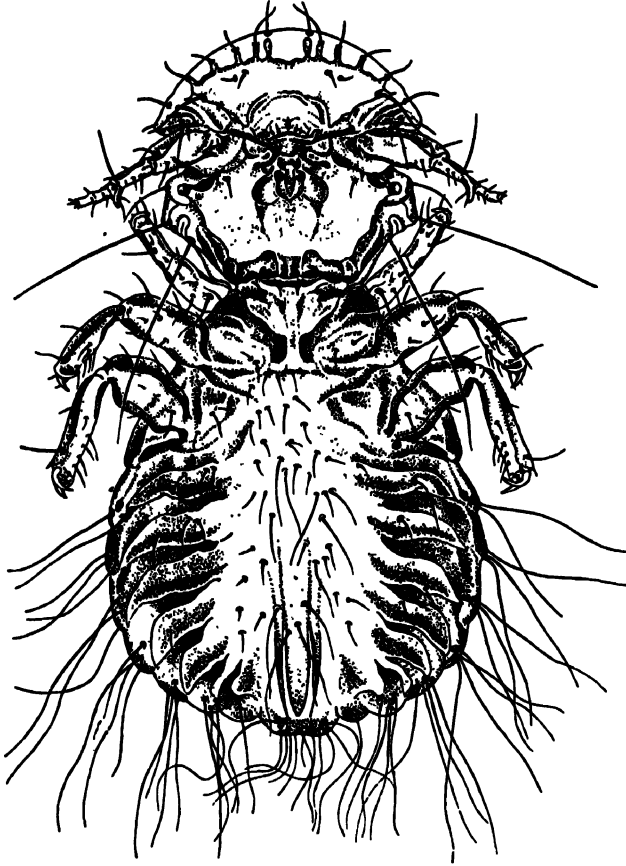


Fig. 31.4. *Goniodes* sp. Chicken louse, male. ×42. (Reis and Nobrega.)

room. The birds are held by the wings and plunged into the dip, leaving the head above the solution. The dip is rubbed into the feathers and skin, and then the head is immersed before holding the bird up to drain for a few moments prior to its release. Rubber gloves may be worn to protect against irritation, particularly if skin wounds are present on the hands. Metallic containers should be rinsed after using.

B. Derris dipping. Freshly powdered derris root, one-fourth ounce is added to each gallon of water and used as a dip.

C. According to Telford (1945b) complete control of chicken lice within 2 days occurred by dipping the birds in a 0.03 per cent water emulsion of DDT.

*Dusting methods for lice.* Materials in powdered form may be applied by the pinch method, by means of a powder shaker, or by dry-dipping. Inhalation of the powder should be avoided by the operator, and skin wounds should be protected by wearing rubber gloves.

In using the pinch method, the operator holds the bird by the wings over a table or a wide, shallow pan. The powder is applied to the skin with a ruffling motion to the feathers. A pinch of powder is placed on each of the following locations: head, neck, back, breast, below the vent, tail, each thigh, and under each wing.

For the powder-shaker method, the bird is held while the powder is shaken on and rubbed into the skin from a can having a perforated top.

Dry-dipping is carried out by holding the bird, back downward, over a wide pan containing the powder. A second operator rubs the powder into the feathers, beginning at the lower parts. The excess powder falls back into the pan.

Substances dusted on birds for lice or used for dry-dipping include the following:



Fig. 31.5. *Columbicola columbae.* Slender pigeon louse, female. ×48. (Reis and Nobrega.)

A. Powdered commercial sodium fluoride. One pound will treat about 100 birds by the pinch method according to Bishopp and Wood (1931). When used in a powder shaker it may be diluted with three times its bulk of flour, powdered talc, or fine dust.

B. Powdered sodium fluosilicate may be substituted for sodium fluoride.

C. Orthophenylphenol may be used by mixing 1 part of it to 4 parts of talc or fuller's earth (Chandler, 1930).

D. Powders containing derris or pyrethrum substances. These are often combined. Powdered pyrethrum flowers 10 to 20 per cent by volume are mixed with powdered derris root so that the final product contains 1 per cent rotenone (present in the derris root). Inert bases such as talc or fuller's earth are added to make 100 per cent volume. This is the usual basis for compounding the various commercial louse powders containing these substances. Such powders deteriorate with age, hence they should be used as fresh as possible. Powdered pyrethrum is said to deteriorate at the rate of about 5 per cent in nine months.

E. Powdered sulfur may be applied from a powder shaker and is effective if relatively large quantities are used.

F. Telford (1945a) reported results of using thirty-seven powdered compounds by salt-shaker application on 149 chickens. Those acting quickly and showing residual properties were (a) DDT, 0.5 to 4 per cent; (b) derris (containing 5 per cent rotenone), 10 per cent; (c) tetramethyl thiouram salts, 2 per cent and 10 per cent; (d) powders containing 30 per cent sulfur mixed with one of the following: Lethane B-71, 5 per cent; Thanite, 5 per cent; Phenothioxine, 5 per cent; NH dust, 5 per cent; Diphenyl, 5 per cent; and Pyrethrins, 0.066 per cent. He also stated that six birds dusted with DDT, 4 per cent, became reinfested in 30 to 34 days after exposure to infested birds.

G. Comparative studies on the efficacy of certain old and new insecticides in the control of lice were made in Hawaii by Alicata, Holdaway, Quisenberry, and Jensen (1946). They stated that 10 per cent Lethane A-70 and full concentration of NH dust were both superior to the other eight substances tried. Also 5 per cent DDT, sodium fluoride, and sodium fluosilicate were effective against body and wing lice but were not as lethal against mites as were the preceding substances.

H. Parish (1942) used phenothiazine on louse-infested chickens. This compound freed the birds within 48 hours, and the residual effect lasted 21 days.

### BEDBUGS AND ALLIED INSECTS AFFECTING POULTRY

The insect order HEMIPTERA includes the true bugs, several species of which parasitize birds by sucking blood. Two families are of particular interest. The family Cimicidae contains the so-called true "bedbugs," and the family Reduviidae includes the "assassin bugs." The latter ordinarily are predaceous on other insects; however, some species do attack man and animals.

The true "bedbugs" are flattened dorsoventrally, thus allowing them to creep into crevices where their young are raised. The adults measure about 2

to 5 mm. in length by 1.5 to 3 mm. in width. The color varies according to species from brown to yellow or red. They have small padlike wing remnants. The suctorial mouthpart structure or "beak," which is jointed, folds under the head and part of the thorax when not in use. Deeply pigmented eyes are prominent on the head. The abdomen has eight segments. Stink glands provide the bug and its surroundings with an unpleasant odor.

The female bedbug lays several 1 mm. sized eggs per day in crevices until about 200 have been deposited. The eggs hatch in about 10 days. There



FIG. 31.6. *Cimex lectularius*. Common bedbug, male. ×15. (Benbrook and Sloss.)

are five nymphal stages, the nymphs feeding at each stage and hiding to digest the meal of blood and to molt their skins. From egg hatching to adulthood requires about 40 days. Nymphs may withstand starvation for about 70 days, and the adults may live for about one year without food. Feeding usually occurs at night, the bugs becoming engorged with blood within 10 minutes.

If attacked by large numbers of bugs, young birds especially may be seriously depleted of blood. The bites are usually followed by swelling and itching due to the injection of saliva into the wound.

The most widespread of the "bedbugs" is *Cimex lectularius* (Fig. 31.6), which attacks man and most other mammals and poultry. It is most prevalent

in temperate and subtropical climates. Poultry houses and pigeon lofts may become heavily invaded.

Other "bedbugs" reported to attack birds include the following species:

*Haematosiphon inodora,* the Mexican chicken bug or "coruco," which also occurs in southern and western United States and in Central America.

*Oeciacus hirundinis* and *O. vicarius,* commonly found in the nests of swallows (particularly barn swallows) whence they may spread to poultry and to man (see Myers, 1928).

Opinions differ as to the geographic distribution of these species of bugs, especially in the United States.

In addition to the above, numerous other species of the true "bedbug" family have been reported on birds in various countries outside the United States.

The insect order HEMIPTERA also includes bugs of the family Reduviidae as previously mentioned. These are usually of minor interest in avian parasitism. "Assassin bugs," "cone-nosed bugs," and "kissing bugs" are common terms applied to such insects, of which there are many species. Only a few of them have learned to suck the blood of mammals and birds. They are larger than true "bedbugs," being up to 25 mm. in length, and they have well-developed wings; otherwise their morphology, life cycles, and behavior are quite similar. Species of reduviid bugs reported as attacking poultry in the United States include *Triatoma sanguisuga* in Maryland, Florida, California, and Texas, and *Triatoma protracta* in Utah and California. It is of interest to note that in 1940, *Triatoma sanguisuga* was found to harbor the virus of equine encephalomyelitis in Kansas by Kitselman and Grundmann (1940). This virus has also been found in natural infections in pigeons and pheasants.

**Control of "bedbugs" and allied insects.** Treatment must be directed at the birds' surroundings during the daytime, because the bugs ordinarily feed at night. In the past, fumigation with hydrocyanic acid gas or with the sulfur dioxide released by burning sulfur was considered to be an effective control method. These fumigants have been principally supplanted by spraying with DDT or pyrethrum extracts.

A. Kulash and Maxwell (1945) used 5 per cent DDT in kerosene to control bedbugs effectively in a chicken house that had been infested for fifteen years. After removal of the birds, the uncleaned house was thoroughly sprayed, followed by removal of litter. No toxic effect on the birds was noticed after they were replaced in the building.

Madden, Lindquist, and Knipling (1945) employed a large number of substances against bedbugs. Of these, only DDT and pyrethrum were satis-

factory; and 5 per cent DDT in kerosene was the most practical when sprayed so as to leave at least 100 mg. per sq. ft., complete kill occurring 48 hours after exposure.

Barnes (1945) tested the residual toxicity to bedbugs of 4 per cent oily DDT solution on cement, unpainted wood, wood painted for years, wood painted for three weeks, and glass. Cement and unpainted wood, bearing 0.23 mg. per sq. cm., yielded a mortality of about 80 per cent after three months and 60 per cent after six months. Wood painted for years supplied about 80 per cent mortality for one month, then toxicity decreased to one-fourth that of cement and unpainted wood. Wood surfaces painted for three weeks were not markedly toxic more than 48 hours after spraying. Glass surfaces with 0.03 mg. per sq. cm. showed more than 60 per cent toxicity after six months.

B. Benzene hexachloride (hexachlorocyclohexane), the active ingredient of which is the gamma isomer, has shown high toxicity to the common bedbug in recently reported, but limited trials (anonymous, 1945). Further work will be necessary before definite recommendations can be made. The musty odor of this compound may be objectionable.

C. Spray compounds, as described later, used against red mites, are also quite effective against bedbugs.

### FLEAS AFFECTING POULTRY

Many species of fleas have been found on birds, but only three or possibly four species have been reported from poultry in this country. However, fleas are very adaptable blood-sucking insects and may attack various host species, especially those closely related. They are cosmopolitan in distribution although more abundant in temperate and warm climates.

They may be recognized as brown to black, laterally flattened insects having the ability to run rapidly along the skin and to propel themselves in the open by leaping. In size they vary from about 1.5 to 4 mm. in the adult stage. The adults suck blood one or more times during the day, although some species are nocturnal. The sticktight flea usually remains attached to the host for days or weeks at a time. Female fleas deposit several eggs per day which roll off the host into surrounding litter where they incubate. Dampness is essential for further development. The eggs are white, almost spherical bodies less than 1 mm. in diameter. Within one to several weeks, depending upon species and climate, the eggs hatch, liberating tiny maggot-like larvae that feed partly on organic matter found in dust and litter, but their principal food is flea feces deposited conveniently by the adult fleas. This flea feces is rich in host blood-products, thus providing a highly nutritious diet for the larvae. After the larvae have grown and shed their skins, usually twice in a period varying from one to several weeks, they proceed

to spin silken cocoons, entangling the thread with various particles of dust and dirt. Then follows the inactive pupal stage for a period varying from one week to months, depending upon the temperature. During this period the pupae transform into white, then yellow, then brown fleas. Emerging from the pupal cocoons, the young fleas seek a host, suck blood, and reach maturity within a few days.

Immature fleas may live for weeks or months without food. Adult fleas may also live for weeks without feeding, but when a host is available their life span may extend over many months to a year or more. The total length of the life cycle is thus seen to vary greatly depending upon such factors as temperature, humidity, exposure, and host availability. For all species it is a matter of weeks at least.



FIG. 31.7. *Echidnophaga gallinacea.* The sticktight or tropical chicken flea, female. Much enlarged. (U.S.D.A., Bur. Entomol. and Plant Quar.)

The fleas of domestic poultry in the United States commonly include the following species: *Echidnophaga gallinacea,* the sticktight flea; *Ceratophyllus gallinae,* the "European" chicken flea; and *Ceratophyllus niger,* the "Western" chicken flea. These will be discussed briefly before flea control is considered.

*Echidnophaga gallinacea* (Fig. 31.7), the sticktight or tropical chicken flea, more often occurs in the southern United States, although occasionally it is found as far north as New York. The adult is about 1.5 mm. long and is reddish brown in color. These fleas usually attach to the skin of the head, often in clusters of a hundred or more. The mouthparts are deeply embedded into the skin so that it is difficult to dislodge them. The adult females forcibly eject their eggs, so that they reach surrounding litter, one to four eggs per day being produced. Incubation takes from 4 to 14 days; the larval period lasts from 14 to 31 days; the pupal period for 9 to 19 days; and the newly emerged fleas mature in from 11 to 18 days.

Sticktight fleas have also been found attacking horses, cattle, deer, dogs, coyotes, foxes, cats, rabbits, skunks, squirrels, rats, and mice. To some extent they attack man, but probably do not breed in human habitations. In addition to chickens, the sticktight flea has been reported from turkeys, ducks, quail, hawks, jays, owls, and English sparrows.

This flea has not been accused of carrying infectious disease agents to chickens. The irritation and blood loss attributed to it may damage poultry seriously, especially young birds, in which death may occur. Production is lowered in older birds. Alicata (1942) experimentally transferred the virus of human endemic typhus from infected rats to guinea pigs through the agency of sticktight fleas, thus indicating a possible public health importance of these parasites.

*Ceratophyllus gallinae* (Fig. 31.8), the "European" chicken flea, also
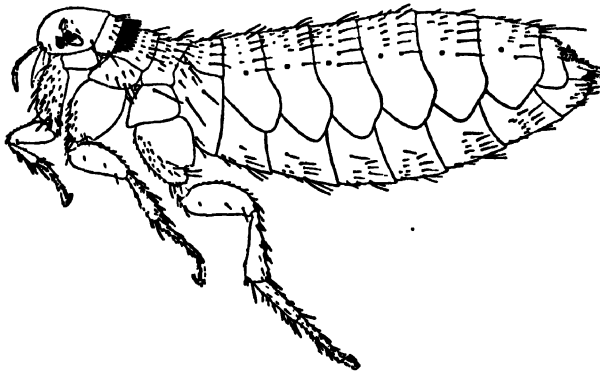


Fig. 31.8. *Ceratophyllus gallinae.* The "European" chicken flea, female. Greatly enlarged. (Reis and Nobrega.)

occurs in the United States. It has been reported from Maine, Massachusetts, Connecticut, New York, Michigan, and Iowa. Undoubtedly, it has a much wider distribution. The hosts include, besides chickens, various other birds, especially English sparrows, bluebirds, and tree swallows, and also man and chipmunks. The adult female measures from 3 to 3.5 mm. in length. This flea behaves like most fleas in that it stays on birds only long enough to feed, its breeding activities occurring in the nests and other surroundings. Otherwise, its effects are like those of the sticktight flea.

*Ceratophyllus niger niger,* the "Western" chicken flea, appears, for the present, to be confined to the Pacific Coast area, although it has been reported from the bluebird in New York. It may attack various mammals, as well as chickens and turkeys, but its principal breeding place is in birds' nests. Grossly and in habits, it resembles *C. gallinae.*

Fox (1940) reports nine additional species of fleas that have been found on birds other than poultry.

**Control of fleas affecting poultry.** This involves not only ridding the birds themselves of fleas, particularly the sticktight flea; but also ridding the nests and housing areas of these parasites. In this respect, flea control methods are quite similar to those used against red mites and bedbugs.

To kill sticktight fleas on chickens, the areas of skin to which the fleas attach may be lightly coated with one of the following preparations, care being taken not to injure the eyes:

A. Sulfur 1 part, petrolatum 4 parts.
B. Phenol 1 part, petrolatum 5 parts.
C. Kerosene 1 part, lard 3 parts.

In heavy infestations of sticktight fleas the birds should also be dipped in one of the following:

A. 2 per cent saponated cresol solution.
B. 2 per cent phenol solution.
C. Laundry soap 1 ounce dissolved in 3 quarts of warm water. As a substitute for dipping, the birds may be thoroughly dusted with 1 part derris powder in 2 parts of talc.

To rid poultry houses of fleas it is suggested that preliminary spraying be done before the houses are cleaned. This will eliminate many fleas, their eggs, and larvae, and will prevent fleas from escaping. Stage (1946) reported excellent control of sticktight fleas by spraying the surfaces and runs of chicken houses with 5 per cent oily solution of DDT, applied at the rate of 1 gallon per 4,000 sq. ft. Emulsions of DDT were equally effective. Other sprays include standard pyrethrum fly sprays, kerosene, light fuel oil, diluted crude petroleum, creosote oil, 2 per cent phenol, or kerosene emulsion. The house cleanings may then be gathered and burned, using additional oil to insure their destruction. A final spraying of the house will destroy those fleas remaining in crevices. If heavily invaded, the houses should again be sprayed in about 10 days. Fumigation with sulfur or hydrocyanic acid gas is very effective against fleas. Because fleas may breed in the dirt beneath buildings, these areas may be heavily soaked with salt water, using about $2\frac{1}{2}$ pounds of ordinary salt (sodium chloride) per gallon of water. About 10 gallons of salt solution should be applied to 100 square feet of earth. The earth under poultry buildings, as well as poultry yards, may be sprayed with a 2.5 per cent aqueous suspension of DDT, at the rate of 1 gallon per 1,000 sq. ft., according to Stage (1946). Poultry, dogs, cats, and rats should be screened away from under buildings, as they may serve to perpetuate flea invasions. Sunlight, hot dry weather, excessive moisture, and freezing hinder the development of fleas; whereas darkness, coolness, dampness, and warmth favor them.

## ADULT BEETLES AND BEETLE LARVAE AS PESTS OF POULTRY

Beetles are included in the insect order COLEOPTERA. They are stoutly armored insects, having two pairs of wings, the membranous hind wings being covered by horny, sheathlike fore wings termed elytra. The adult female beetle lays eggs from which hatch larvae, commonly called grubs,

followed by a pupal stage. The adults and larvae show great variation in habits. Some live on land, others in water. Certain species feed on animal matter, others on plants; thus many species are useful as scavengers or in reducing other insect populations. Numerous species cause considerable harm by destroying plants and plant products or by acting as pests to animals. Other species serve as intermediate hosts for internal parasites.

Some of the beetles more important to man and his animals are listed under the common names of scavenger beetles, dung beetles, ground beetles, grain beetles, larder beetles, wood borers, June beetles, potato beetles, Japanese beetles, weevils, mealworms, ladybirds, leaf chafers, and blister beetles.

As far as birds are concerned, certain adult beetles and their larvae may act as pests or may serve as intermediate hosts for internal parasites, particularly certain of the tapeworms. Fourteen species of beetles are known to transmit the following tapeworms of chickens: *Raillietina cesticillus, Choanotaenia infundibulum, Hymenolepis carioca,* and *Hymenolepis cantaniana.* Because beetles make up part of the diet of birds and because some beetles eat avian carcasses, they may transmit bacterial or virus infections; but of this little is known.

Beetle larvae acting as pests of poultry include the following species:

*Tenebrio molitor,* the yellow mealworm, is ordinarily found in the adult and grub stages consuming grain products stored in mills, warehouses, bakeries, and groceries. The beetles are shiny brown to almost black in color and about 15 mm. long. They may infest setting hens, attacking mainly the feet where the loss of skin may be followed by severe hemorrhage. The larvae or grubs, known as flour, meal, or bran "worms," are smooth, hard, yellow, cylindrical, wormlike creatures about 30 mm. long. These grubs have been found to erode the skin of young pigeons. According to Levi (1941) other related mealworm beetle larvae may produce similar damage.

*Dermestes lardarius,* the larder beetle, and related species, ordinarily destroy stored grain products and meats (especially ham and bacon) or feed on hides, skins, furs, museum specimens, or decaying animal matter, notably the accumulated droppings in pigeon lofts. The adult larder beetle is about 7 mm. long, black in color, and the basal half of each wing cover is brownish yellow crossed by a band of three black spots. The larvae are about 12 mm. long, dark brown above, gray below, and are covered with brown hairs. The larvae may attack the skin of nestling pigeons.

*Silpha thoracica, Silpha apaca, Necrophorus vestigator,* and possibly other species of the beetle family, Silphidae (carrion beetles) may also breed in pigeon droppings. The grubs, which are about 15 mm. long, and black,

are reported to invade the skin of squabs, and the wounds produced may be secondarily infested by fly maggots.

**Control of beetle larval invasions.** This is based upon cleanliness of houses or lofts, from which droppings should be removed frequently. Grain and feed storages and hides and skins should be kept away from birds. If infested, such substances should be fumigated or otherwise treated according to directions that may be obtained from the United States Department of Agriculture. Wounds produced by beetle larvae should be gently cleaned, using commercial benzol to destroy the grubs, followed by daily irrigation with a mild disinfectant solution until healing occurs. Reporting on the control of bedbugs in chicken houses, Kulash and Maxwell (1945) found that a 5 per cent solution of DDT in kerosene also killed adult dark mealworms, *Tenebrio obscurans*.

*Macrodactylus subspinosus*, the rose-chafer, is a leaf-chewing beetle that, according to Lamson (1922), is highly poisonous when ingested in quantity by chicks, ducklings, goslings, poults, and young game birds. These beetles are found mainly in the regions of the Atlantic coast, central states, and Middle West. Symptoms of poisoning appear about 4 to 5 hours after ingestion. The birds become sleepy, the wings droop, and muscular weakness develops. Death may occur in from 5 to 24 hours. The condition is diagnosed by finding the beetles in the crop. No specific treatment for affected birds is available, although a saline laxative is advisable. Plants infested with rose-chafers may be sprayed with lead arsenate solution.

## MOSQUITOES AFFECTING POULTRY

Although mosquitoes are not as important to poultry as they are to man and other mammals, they are of some direct and indirect interest. Some eighty species have been described from North America. How many of these suck blood from poultry is not known.

Mosquitoes are recognized as two-winged insects belonging to the family Culicidae of the order DIPTERA. Most species are about 5 mm. in length, and the wings are characteristically veined and scaled. The legs and the abdomen are long and slender, and the small spherical head of the female is provided with elongated mouthparts for piercing the skin. The male does not suck blood, but does live from plant juices, nectar, and other fluids.

Mosquitoes deposit their eggs on pools of water in which the larval and pupal stages are passed. The adults emerge from the pupal cases, quickly breed, and then seek a host. In warm weather the life cycle is completed in about 7 to 16 days for the more common species. The adults are most active on quiet days, especially toward evening.

That mosquitoes may attack poultry in swarms is stated by Bishopp

(1933), who reported the deaths of numerous chickens in Florida. The offending species was *Psorophora columbiae*. Besides blood loss, the birds appeared to show toxicity from the bites.

Fowl pox is transmitted by the mosquitoes *Aedes stimulans, A. aegypti,* and *A. vexans* according to Brody (1936) and also Matheson *et al.* (1931). The first-named species harbored the virus for 2 days, whereas the last-named species continued to infect birds up to 39 days after contacting the virus of fowl pox and pigeon pox. Mosquitoes also transmit one type of avian malaria (*Plasmodium* sp.) according to Herman (1938a). For the most part the birds affected are the smaller wild species including canaries. Recently Davis (1940) has called attention to the relationship of birds and mosquitoes as hosts for the virus of eastern equine encephalomyelitis.

Mosquito control methods, as applied to dwellings and other buildings and their surroundings, may be used to equal advantage in and around poultry houses and poultry raising areas. Oily and aqueous and aerosol sprays, or dusts containing DDT alone, or DDT with pyrethrum, or containing other of the newer organic compounds, may be employed by following the directions supplied by the manufacturers. Preventive measures by screening and the use of repellants are of doubtful practical value in poultry husbandry.

## FLIES AND GNATS AFFECTING POULTRY

The insect order DIPTERA includes the two-winged forms, not only the mosquitoes but also many families of flies and gnats. Certain species of these groups may cause harm to poultry in several ways: by sucking blood, by injecting toxic substances, by acting as intermediate hosts for certain tapeworms, by harboring botulinus toxins, or by larval invasion of body openings and skin wounds.

*Pseudolynchia canariensis* (Fig. 31.9), the pigeon fly, louse-fly, or flat fly is a rather important parasite of domesticated pigeons in warm or tropical areas. It has been known since 1896 in the southern half of the United States and also occurs in many other countries.

The adult fly is dark brown and about 6 mm. in length. The two transparent wings are somewhat longer than the body. These flies move rapidly through the feathers, and they suck blood, particularly from nestling pigeons about two to three weeks of age. They may also bite man, inflicting a painful skin wound that persists for several days.

The female pigeon fly deposits on the birds her white larvae, about 3 mm. in length, enclosed in a pupal case. This rolls off the host into the nest and in a few hours the pupal case hardens and turns black in color. After a pupal stage of about 30 days, the fly emerges, living for about 45 days, during which time the female deposits four or five young.

Infested pigeons suffer from blood loss and from irritation. Also, the pigeon fly may transmit a protozoan blood-cell parasite, *Haemoproteus columbae,* the cause of a malaria-like disease of pigeons.

There are several other louse-flies related to the pigeon fly that infest various species of wild birds, notable among which is the fly *Lynchia hirsuta,* a transmitter of *Haemoproteus lophortyx,* the cause of California valley quail malaria.
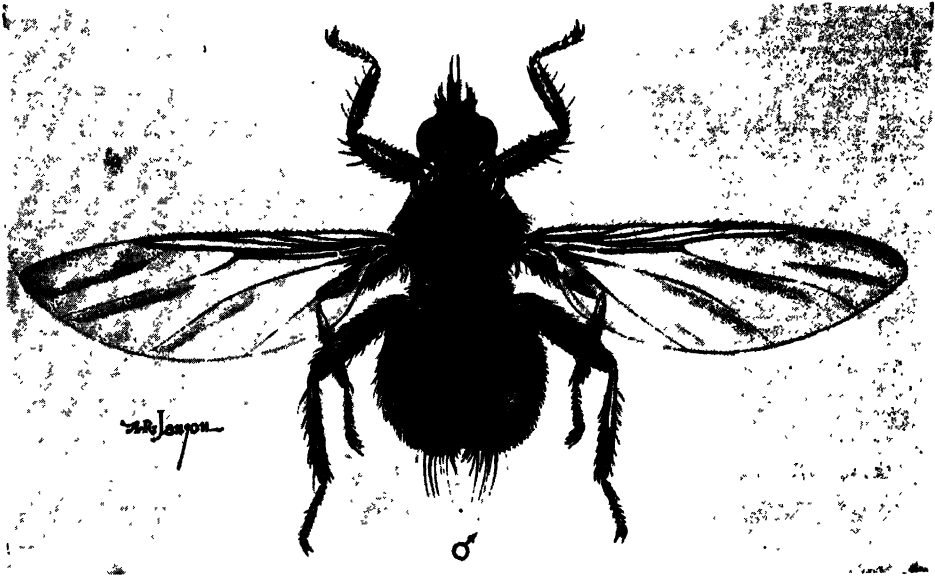


FIG. 31.9. *Pseudolynchia canariensis.* The pigeon fly, louse-fly, or flat fly. Enlarged. (Drake and Jones.)

**Control of the pigeon fly.** Because these flies breed in pigeon nests, it is essential to clean the nests and surroundings at 15- to 20-day intervals and to burn or bury the cleanings. Pigeon lofts may be rid of adult flies, according to Levi (1941), by using a pyrethrum-containing fly spray (1 part pyrethrum extract to 2 parts of kerosene). This should not be sprayed on unhatched eggs. Pigeons may be freed from the flies by applying to them several pinches of fresh pyrethrum or derris powder, rubbing it into the skin. Tobacco dust (containing 6 per cent nicotine) may similarly be used. Dipping may be resorted to, in which case derris powder one-half to 2 ounces is added to 1 gallon of soft water containing 2½ ounces of laundry soap.

**Black flies.** These dipterous insects, variously called turkey gnats and buffalo gnats (Fig. 31.10), belong to the fly family of Simuliidae. They are blood suckers, of which more than fifty species are known. At times they may cause serious damage to man and to livestock. Their importance to

poultry raisers is that they may attack in swarms, depleting the birds' blood volume and injecting toxic material. They also transmit certain blood protozoa belonging to the genus Leucocytozoon.

Black flies are tiny, hump-backed, two-winged flies from about 1 to 5 mm. in length and black or nearly black in color. The females are vicious blood suckers during daylight hours. They breed in running or slowly moving water from which they may travel several miles in search of blood. Eggs are laid on solid objects at the edge of water. The larvae emerge in from 5 to 30 days and enter the water, attaching to stones or other objects. After about three to ten weeks, during which the larvae molt six times, the pupal stage is reached. This stage, too, occurs under water, lasting from a few days to a month. The adult flies emerge during warm weather. Hibernation occurs in the egg or larval stage.

Simuliids are widely distributed, but they occur mostly from the north temperate to the subarctic regions. Reports of their occurrence in this country date

FIG. 31.10. *Simulium* sp. One of the black flies, buffalo gnats, or simuliids. Enlarged. (Iowa State College.)

back to the early part of the last century when buffalo gnats seriously interfered with homesteading operations in the South. It was then noted that they would swarm on poultry, forcing setting chickens and turkeys to leave their nests, and they killed young birds by forcing their way in large numbers under the wings where they sucked blood.

Walker (1927) reported that *Simulium bracteatum* fatally attacked goslings in Canada, and Gibson (1930), also of Canada, found that *Simulium* sp. caused losses to chickens and turkeys. Underhill (1939) stated that *Simulium nigroparvum* and *S. slossonae* attacked turkeys in Virginia, as far as fifteen miles away from their breeding places. In 1928 heavy losses in chicks occurred in western Iowa. Swarms of gnats produced severe anemia, leaving a hemorrhage at each skin area punctured. The chicks ingested enormous numbers of the gnats so that their crops were distended with them.

It was not until 1932 that disease transmission by gnats to poultry was proven. Skidmore (1932a), of Nebraska, found that *Simulium occidentale* could transmit *Leucocytozoon smithi,* a blood protozoan of turkeys. O'Roke (1934) showed that *Simulium venustum* transmitted *Leucocytozoon simondi* to tame and wild ducks in Michigan.

**Black fly control.** This is difficult because these pests breed in streams containing rocks, brush, and logs. Stream clearance may help, but it is an expensive procedure. Sweeping the downstream faces of dams may dislodge

many pupae. The drifting smoke of smudge fires will repel the adult flies. Birds may be kept within screened enclosures during the daytime, using screen of 24 mesh per inch or smaller. Repellents are helpful, for example, oil of citronella 1 part in light mineral oil 4 parts may be sprayed on the outside feathers. Dove (1945) quoted Hutson's report on the use of 1 per cent DDT dust for the control of the black fly, *Simulium venustum*, on golf course greens, tees, and shrubbery in Michigan. A liberal dusting kept the premises practically free of flies for approximately one week.

*Musca domestica*, the common house fly, is frequently eaten by birds. It is an intermediate host for three species of poultry tapeworms, namely: *Raillietina tetragona* of the chicken, turkey, guinea-fowl, and quail; *Raillietina cesticillus* of the chicken, turkey, and quail; and *Choanotaenia infundibulum* of the chicken, turkey, duck, and pigeon. Skidmore (1932b) reports that common house flies that have fed on infected fowl cholera blood can transmit this disease when fed to turkeys.

*Stomoxys calcitrans*, the blood-sucking stable fly, attacks most mammals and birds. Fortunately, the bite is not poisonous, although such areas may become infected, and the blood that oozes from bites may attract maggot-producing flies. The stable fly is an intermediate host for *Hymenolepis carioca*, a tapeworm of the chicken, turkey, and quail.

**Myiasis of poultry.** Invasion of birds by fly larvae (maggots) is not as common as in mammals. Knipling and Rainwater (1937) mention that *Cochliomyia americana*, the primary screwworm fly, will deposit eggs in wounds on chickens, turkeys, and geese. Maggots hatching from these eggs actively destroy living tissue. Stewart (1929) reported a case of cloacal invasion of a hen by screwworm larvae. Invaded wounds may be treated with Smear No. 62, developed in the United States Department of Agriculture and reported by Melvin, Smith, Parish, and Barrett (1941).

The nests of wild birds may become infested by the maggots of various species of flesh and blow flies, with disastrous effects on the nestlings.

Certain fly larvae are of interest because, by breeding on decomposing carcasses, they may ingest toxins of the bacterium *Clostridium botulinum*, according to Bishopp (1923). If poultry eat such maggots, the disease botulism may occur. This has been called "limberneck," a term descriptive of one symptom but not characteristic of botulism alone. The larvae of the following species of flies have been incriminated as transmitters of botulinus toxins, Types A and C: *Lucilia caesar* and *L. sericata* (blow flies); sarcophagid larvae (flesh flies); and *Cochliomyia macellaria*, the secondary screwworm fly. Prompt burial, burning, or other sanitary disposal of animal carcasses will do much to prevent botulism from these sources.

Schalk (1928) noted that "fly larvae" developing on tuberculous chicken carcasses could transmit this disease when fed to nontuberculous chickens.

## MITES OF POULTRY

Numerous and important species of mites may affect domesticated and wild birds. Mites belong in the invertebrate animal group (phylum) **ARTHROPODA**, in the class **ARACHNIDA**, and with the ticks in the order ACARINA. For classifications of mites, reference may be made to Banks (1915) and Ewing (1929a).

Parasitic mites are microscopic or barely visible to the unaided eye. The main body parts include a usually unsegmented, soft abdomen broadly continued anteriorly by a combined head and thorax (cephalothorax) to which the legs are attached. Some mites breathe through tracheal tubes, others by absorption of oxygen through the soft skin.

The typical mite life cycle consists of the egg, the larva (six-legged), the nymph (eight-legged but sexually immature), and the adult (eight-legged). The cycle, in general, takes from one to four weeks for completion, depending upon species, climate, and availability of a suitable host.

Most mites of birds use blood or lymph for food, hence anemia is a more or less constant symptom. It might be expected that blood-sucking mites could easily transmit bacterial infections. The common red mite, *Dermanyssus gallinae,* has been reported as a transmitter of fowl cholera by Hertel (1904) and by Plasaj (1925); and of the fowl spirochete, *Treponema anserinum,* by Hart (1938). The same mite from chickens has been shown by Sulkin (1945) to harbor the virus of equine encephalomyelitis (western type); also Reeves *et al.* (1947) recovered this virus from the northern feather mite, *Liponyssus sylviarum,* obtained from the nests of English sparrows and yellow-headed blackbirds. Smith, Blattner, and Heys (1944) isolated human St. Louis encephalitis virus from *Dermanyssus gallinae.*

Those mites that move rapidly over the skin will irritate birds to a considerable degree. Others burrow into the epithelium, causing tissue proliferation and scab formation. Feather loss results from invasion of feather follicles by certain species, the feather bases being destroyed, or the birds may pull out the affected feathers. Although mites are considered ordinarily to be external parasites, at least two species invade the subcutis or the internal organs of birds.

*Dermanyssus gallinae* (Fig. 31.11), the red or roost mite, is probably the commonest and most widespread of all the mites of birds. Because it breeds in the birds' surroundings, attacking mostly at night, it is apt to be overlooked. The adult female measures about 0.69 by 0.4 mm., varying in color from gray to deep red depending upon its blood content. Chickens are the

commonest hosts, but turkeys may be attacked when placed in infested chicken houses. English sparrows frequently transmit this parasite because of the habit of lining their nests with chicken feathers.
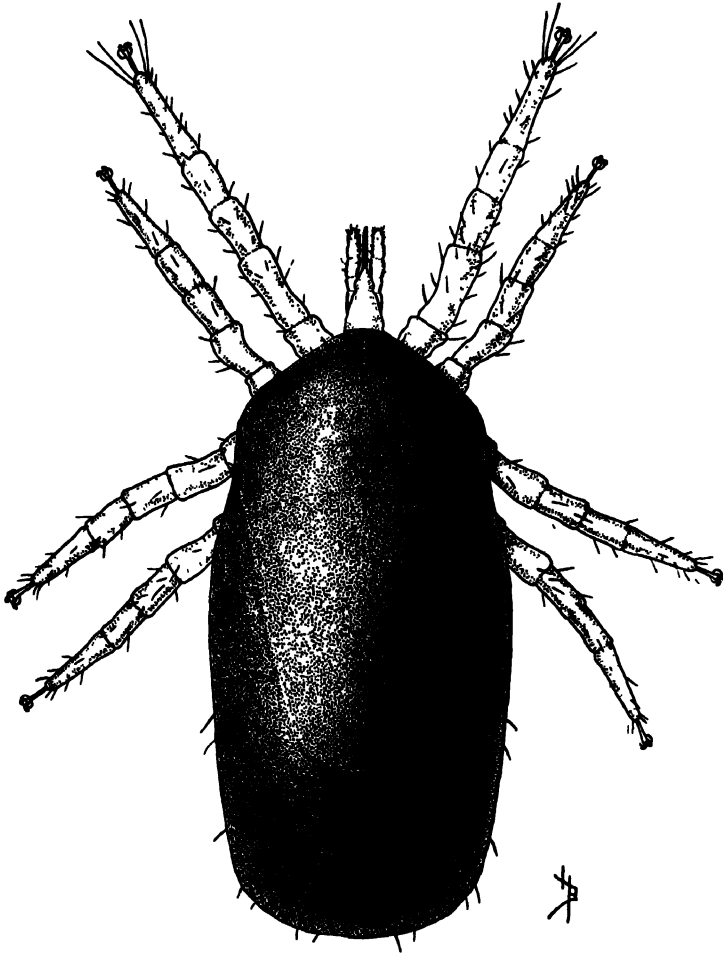


FIG. 31.11. *Dermanyssus gallinae*. The red or roost mite. female. after feeding. Greatly enlarged. (U.S.D.A. Bur. Entomol. and Plant Quar.)

Red mites may live for several months without food. When hosts are available they may not only produce anemia, thereby seriously lowering production, but may actually kill birds through extraction of blood. This is particularly true of young birds and of setting and laying hens. Birds in production may refuse to lay in infested nests. This symptom indicates that poultry houses should be examined for mites.

Control of red mites is best done at weekly intervals by removing birds from infested houses and forcefully spraying all litter, roosts, and crevices.

Then the litter should be removed and either burned or buried. Nests should be flushed with scalding water, thus avoiding chemical odors that might taint eggs. Sprays of value in killing red mites include:

A. Anthracene oil wood preservative (Carbolineum) diluted with an equal part of kerosene.

B. Coal-tar cresol disinfectants in 10 per cent dilution with water.

C. Kerosene emulsion, 10 per cent solution. This is made by shaving one-half pound of laundry soap in 1 gallon of hot, soft water; when dissolved, remove from the fire and add 2 gallons of kerosene; stir to emulsify; add 17 gallons of hot water and stir.

D. Alicata *et al.* (1946) reported that a single application of 10 per cent "Lethane A-70" to a group of twelve chickens infested with common red mites, plus dusting of the house and ground underneath, eliminated all the mites on the birds and reduced to a small proportion the mite population on the houses and on the ground.

E. Steward (1947) reduced considerably the red mite population of heavily invaded



Fig. 31.12. *Liponyssus sylviarum.* The northern feather mite, female. ×73. (Benbrook and Sloss.)

hen houses by using benzene hexachloride 1:5,000 spray. A concentration of 1:2,500 almost eliminated the mites. Dusting the perches and the birds with 0.5 per cent benzene hexachloride was also recommended.

To prevent red mite infestation, inspect houses frequently; quarantine new birds until inspected; disinfest old houses before admitting birds; and destroy nearby sparrow nests if possible.

*Liponyssus sylviarum* (Fig. 31.12) is the northern feather mite, so-called because it occurs mostly in temperate and north temperate as well as in subtropical areas. It has been reported from twenty-two species of birds, also from rats and accidentally on man. It resembles the common red mite, but differs from it in that it occurs both on birds and on their surroundings more or less continuously, even during the daytime. When infested birds are

handled, the mites quickly crawl over the examiner's hands and arms. Parting the feathers reveals the mites, their eggs, cast-off skins, and excrement on the body surface and feathers, making the bird appear soiled. Feather mites are vicious blood suckers. They also cause scabs to form thus injuring the appearance of dressed poultry (Payne, 1930).

*Liponyssus canadensis* is listed by Hearle (1938) as occurring on fowl in Canada, but not being of serious importance.

Control of feather mites involves both the birds and their surroundings. Houses should be sprayed and cleaned as was suggested for red mites. The infested birds may be treated by one of several methods:

A. Dipping in a mixture of warm water 1 gallon to which has been added 1 ounce of laundry soap and 2 ounces of sulfur. This mixture must be kept agitated during use.

B. Nicotine sulfate, 40 per cent solution, may be used as a fumigant, applying it to the roosts as suggested for the control of lice (p. 722). Repeat the application at 3-day intervals for at least three times (Payne, 1929; and Cutright, 1929).

C. In case dipping is not advisable because of cold weather, the birds may be greased around the tail and vent regions with a mixture of powdered naphthalene 1 part in petrolatum 2 parts. In very cold weather the naphthalene may be replaced by paradichlorbenzene which keeps the mixture more liquefied.

D. Dusting with finely powdered sulfur may also be helpful during cold weather. According to Povar (1946) finely powdered sulfur was not found observably toxic to the mites, but it acted as a repellent. He also observed that 10 per cent DDT powder dusted on birds, though somewhat toxic to the mites, did not prove sufficiently effective to replace treatment with nicotine sulfate.

*Liponyssus bursa* (Fig. 31.13), the tropical feather mite, is closely allied to the northern feather mite. It is more prevalent in warm or hot climates although it has been found in the northern United States. Control is accomplished in much the same manner as for the northern feather mite, particular attention being paid to the birds themselves and their nests. The latter should be scalded with water before being cleaned.

*Eutrombicula alfreddugesi,* the chigger, is the six-legged larval stage (Fig. 31.14) of a mite that may infest the skin surface of various bird hosts as well as mammals, including man. The adults are not parasitic. Unfed chigger larvae are from 0.1 to 0.45 mm. in diameter, hence hardly visible unless they are engorged with blood when they appear as minute red dots. The adults breed on the ground, especially along fence rows or in undis-

turbed wooded or brushy areas. The larvae attach to the skin, often in groups, by means of their mouthparts and inject a highly irritant substance into the wound during the blood-sucking period. Itching vesicles or even abscesses may form at the points of attachment, surrounded by a zone of hyperemia and edema. Apparently a toxemia may occur as is indicated by the mortality that follows infestation of chicks, especially quail.

The control of chiggers is difficult if birds are on free range, hence the desirability of keeping young birds in clean open pens especially from June
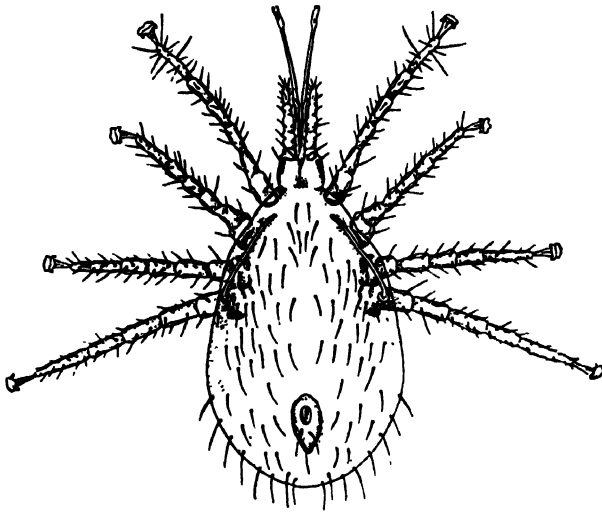


Fig. 31.13. *Liponyssus bursa.* The tropical feather mite, female. Enlarged. (Reis and Nobrega.)

to October. Treatments of individual birds have been recommended as follows:

A. As a preventive the birds should be dusted frequently with finely powdered sulfur. This may also be dusted into the nests.

B. Sulfur ointment may be applied to the skin lesions. This consists of 1 part of sulfur to 5 parts of petrolatum.

C. Balsam of Peru may be applied to lesions.

D. A 5 per cent solution of phenol may be swabbed on the lesions, followed by tincture of iodine. This treatment is recommended if vesicles or abscesses form.

E. Dusting the ground with elemental sulfur or with DDT 1 per cent in pyrophyllite resulted in a reduction of over 80 per cent of the chiggers, according to Smith and Gouck (1944), working in Georgia and South Carolina. The sulfur was dusted at the rate of 96 pounds per acre, and the 1 per cent DDT at the rate of 125 pounds per acre.

*Neoschöngastia americana* is a mite closely related to the chigger. Ewing (1929b) states that it occurs on chickens in the southern United States and that it is very injurious.

*Cnemidocoptes mutans* (Fig. 31.15), the scaly-leg mite is of common occurrence on various birds, particularly the older ones that should ordinarily be culled from flocks. The mites are almost spherical in shape, short-legged, and adult females are about 0.5 mm. in diameter. The male is less than half the size of the female, and the legs are longer. Lesions are produced on the unfeathered portions of the host's legs and occasionally on the skin of the comb and wattles. Tunnels are bored into the epithelium, causing proliferation and the formation of scales and crusts (Fig. 31.16). This type of mite invasion of birds corresponds to sarcoptic mange of mammals. Affected birds may be crippled if the infestation is severe. The mites pass through their entire life cycle in the skin. Transmission to uninfested birds progresses slowly by contact with those infested and with their surroundings.



Fig. 31.14. *Eutrombicula alfreddugesi* larva. The chigger. ×130. (Benbrook and Sloss.)

Control of scaly-leg mites should begin by culling or by isolating the affected birds. Additions to the flock should be inspected for lesions. Houses should be cleaned frequently, especially the roosts, which should be sprayed as recommended for red mites.

Many methods of treatment have been suggested, most of them based upon the application every 3 or 4 days of some oily compound that will penetrate the epithelium:

A. Soak the shanks in soapy water with the aid of a hand brush. Allow them to dry, then apply oil of caraway 1 part in petrolatum 5 parts.

B. Phenol ointment, 2 per cent strength, or sulfur ointment 15 to 20 per cent, may be applied to the legs. The latter is of value for lesions that may occur on the comb, wattles, or neck.

C. An inexpensive flock treatment consists of dipping the shanks in a mixture of kerosene 1 part in raw linseed oil 2 parts. This must not be applied to the feathered areas of the skin.

D. Crude petroleum may be applied every 30 days.

E. Balsam of Peru undiluted or else 1 part in alcohol 3 parts may be used.

F. Kerosene 1 part mixed with lard 5 parts may be applied every other day.

G. Beechwood creosote 1 part in lard 20 parts is effective.

*Cnemidocoptes gallinae,* the depluming or body mange mite, resembles the scaly-leg mite in general structure although it is smaller, the adult female being about 0.3 mm. in diameter. It invades the feathered areas of the epidermis of chickens, pigeons, and pheasants, especially around the feather bases. Intense irritation induces the host to pull out body feathers. The mites are more prevalent in the spring and summer at which time the infestation may spread rapidly by contact. Depluming mites produce injury by interfering with the control of body heat. Some of the affected birds will lose weight and show lowered production.



FIG. 31.15. *Cnemidocoptes mutans.* The scaly leg mite, gravid female. ×150. (Benbrook and Sloss.)

Control is not easily accomplished. Prompt isolation of affected birds and disinfestation of houses as recommended for red mites should come first. Treatments recommended for individual birds include:

A. Dipping in sulfur 2 ounces, soap 1 ounce, and warm water 1 gallon. This mixture should be thoroughly soaked into the skin and especially into the feet of cockerels. If lice are also present, there may be added to the dip, sodium fluoride or sodium fluosilicate 1 ounce.

B. Ointments may be used, consisting of sulfur 1 part, petrolatum 4 parts; or caraway oil 1 part, petrolatum 5 parts.

C. Moisten affected areas with soapy water, then apply powdered pyrethrum or powdered sulfur with the aid of a powder blower.

*Cytoleichus nudus* (Fig. 31.17) is a mite that has learned to live as an

internal parasite of the respiratory system, including the bronchi, lungs, air sacs, and the bone cavities connected therewith in birds. Air-sac mites have been found in chickens, turkeys, pheasants, and pigeons from many parts of the world. Although not of common occurrence, these mites are often overlooked because of their small size and peculiar habitat.

The adult female mites are whitish specks, measuring about 0.5 to 0.6 mm. in length by about 0.4 mm. in width. No details are known of the life cycle, although the usual speculation is that the mites lay eggs in the lower air passages, that these eggs are coughed up and probably are swallowed, reaching the ground in the droppings. The mode of infection is not known.

There is considerable conflict among observers as to the damage done by air-sac mites. Some are of the opinion that they are practically harmless because their presence has been noted in apparently healthy birds. Others state that the mites are responsible for emaciation, peritonitis, pneumonia, obstruction of air passages, and that they are predisposing factors for tuberculosis. Heavy invasions have definitely been associated with grave loss in weight and in weakness, so that the affected birds resemble clinical cases of tuberculosis.

Close inspection of the opened carcass of an affected bird soon after death will show whitish dots slowly moving over the transparent air-sac surfaces. Identification may easily be made by placing



FIG. 31.16. Lesions produced by *Cnemidocoptes mutans*, the scaly leg mite. (Benbrook and Sloss.)

mites in a drop of water on a slide, applying a cover glass, and examining under magnification of 100 diameters.

Little information has been published as to the control of air-sac mites. Most writers recommend the destruction of the carcasses of affected birds, followed by disinfection and cleaning of the poultry house.

*Laminosioptes cysticola* (Fig. 31.18), the subcutaneous or flesh mite, is another example of an originally external parasite invading deeper tissues. It has been reported mainly from chickens, also from turkeys, pheasants, geese, and pigeons in many parts of the world.

Perhaps it normally is a parasite of the surface or upper layers of skin cells. However, it is most frequently noticed in the loose subcutaneous connective tissue. It has even been reported as occurring in the muscles, abdominal viscera, lungs (pigeons), and on the peritoneum.

Ordinarily, subcutaneous mites do not appear to influence the health of infested birds, although the lesions produced may make carcasses unpalatable as food for man.

The female mite measures about 0.25 to 0.26 mm. long by about 0.11 mm. wide. A distinctive feature is the transverse constriction around the body posterior to the second pair of legs. The life cycle is unknown except that the female lays embryonated eggs. Neveu-Lemaire (1938) states that the mite will pass through all stages of its development even in the deeper tissues of the host.

Attention is most often called to subcutaneous mites by the occurrence of yellowish nodules up to several millimeters in diameter in the subcutis. These areas are often mistaken for tuberculous lesions. The nodules appear to be caseo-calcareous deposits formed by the bird so as to enclose the mites after they die in the tissues. Large numbers of nodules are most often found in aged emaciated birds. Kasparek (1907) reported L. cysticola in pigeons in which the mites were surrounded by nodules in the lungs, causing death.



FIG. 31.17. *Cytoleichus nudus.* The air-sac mite, male. ×100. (Benbrook and Sloss.)

Perhaps more careful examination of the skin and subcutis of birds under a dissecting microscope might reveal the presence of this parasite more frequently. Otherwise, diagnosis will depend upon finding the characteristic nodular lesions and by seeing the mites or their remains in nodules that have been crushed under a cover glass in a drop of water.

Apparently no attempt has been made to control subcutaneous mites except by the destruction of affected birds.

*Epidermoptes bilobatus* (Fig. 31.19) is a skin mite frequently reported from Europe and more rarely from South and North America, according to James *et al.* (1930). It occurs on chickens and apparently may or may not produce lesions. The adult female is about 0.17 to 0.22 mm. long. When lesions are produced, they consist first of a fine scaly dermatitis. This may be followed by the formation of thick, brownish, sharply-edged scabs. Neveu-Lemaire (1938), of France, suggests that the more severe lesions may be due

partly to a concomitant fungous infection by *Lophophylon gallinae*, also that birds affected with scaly-leg mites often have depluming mites at the same time. Epidermoptic scabies may at times result in emaciation and even death.

Treatment of infested birds is recommended as for depluming scabies. In addition, Neveu-Lemaire (1938) suggests the use of Balsam of Peru and alcohol, equal parts, applied to the skin.

*Rivoltasia bifurcata*, a feather-eating mite similar to *Epidermoptes*, has been reported on chickens in Europe, by Reis (1939) in Brazil, and by Bushnell and Twiehaus (1939) in a Kansas publication. In size it is said to be 0.25 by 0.15 mm. Only slight damage to feathers has been noted.

*Syringophilus bipectinatus* is commonly known as a quill mite. Originally described in Europe in 1880, it was not until 1932 that it was reported in the United States by Rebrassier and Martin (Ohio) from the chicken, turkey, and golden pheasant. A similar mite, *S. columbae*, has been described from pigeons by Hirst (1922). Wild birds harbor related species.
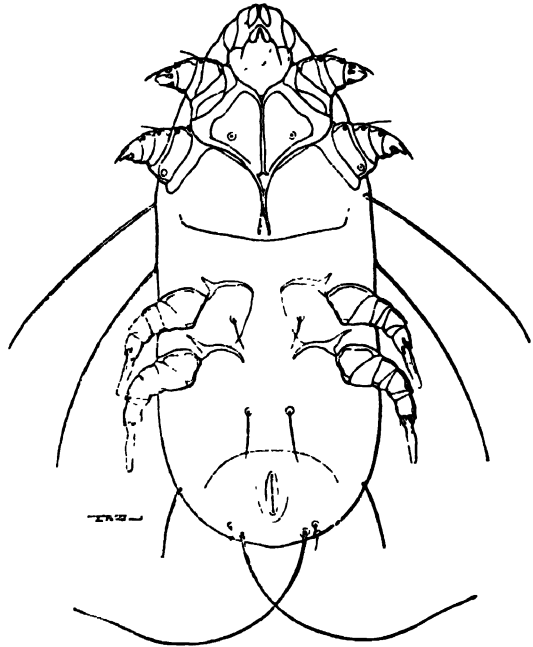


FIG. 31.18. *Laminosioptes cysticola*. The subcutaneous mite, male. ×376. (Reis and Nobrega.)

*S. bipectinatus* females measure up to 0.9 mm. in length and to 0.15 mm. in width. The mites appear to cause partial or complete loss of feathers. The remaining quill stumps contain a powdery material in which the mites may be detected under low power magnification. No specific method for control has as yet been described. It would appear advisable to dispose of affected birds, then disinfect and clean their quarters.

*Falculifer rostratus* (Fig. 31.20), another feather-damaging mite, occurs principally between the barbs of the large wing feathers of pigeons, according to Pillers (1927). Although reported mainly in Europe, this mite has been noted in the United States, and it may be more or less widespread. In

size the mite is only about 0.5 mm. long. Pillers (1927) believes that the feathers are injured by the loss of barbules, whereas Levi (1941) states that he has not found the feathers to be harmed. There is some evidence that the nymphal stage of the mite may occur in the subcutis or the internal organs.
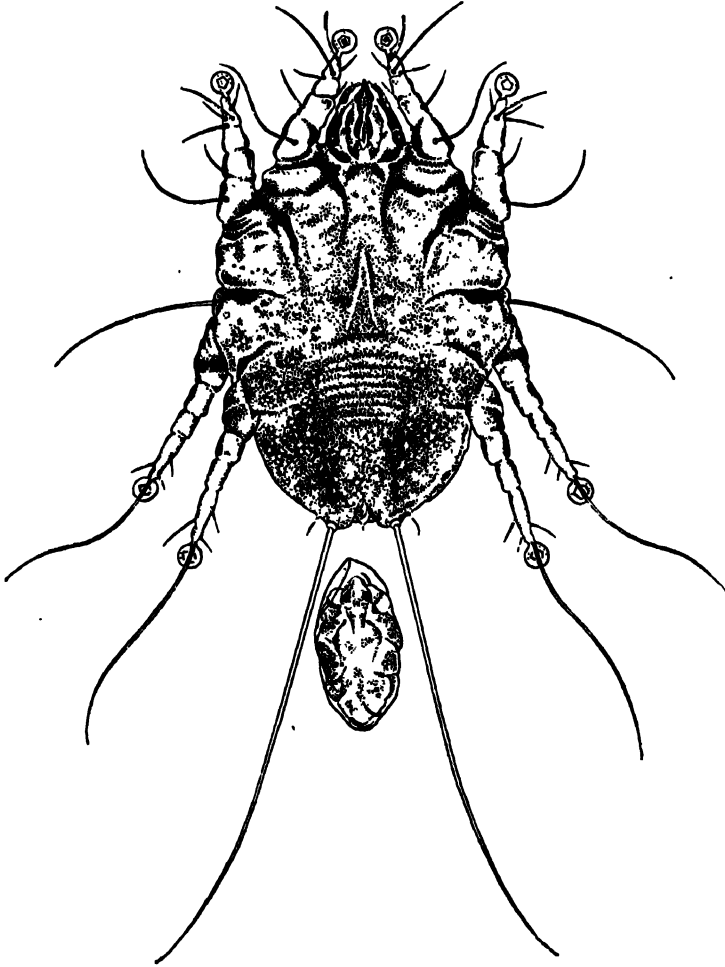


FIG. 31.19. *Epidermoptes bilobatus*. The epidermoptic scabies mite, female. ×200. (Reis and Nobrega.)

Pillers (1927) and others recommend that infested pigeons be fumigated with sulfur dioxide gas. This appears to be a tedious and rather dangerous procedure. Probably a better method would be the use of a sulfur-containing dip such as that recommended for *Liponyssus sylviarum,* the northern feather mite.

*Freyana chaneyi,* also a feather-inhabiting mite, has been reported from

turkeys in Maryland by Chapin (1925). Its prevalence in Texas and Louisiana is mentioned by Bushnell and Twiehaus (1939). These mites congregate in the grooves on the under sides of the shafts of the wing feathers.

*Megninia gallinulae* is a rarely reported mite according to Wickware (1921) in Canada. Apparently it is associated with loss of scales from the lower legs of chickens and with a crusty dermatitis in the head region. Neveu-Lemaire (1938) lists a similar species, *M. cubitalis*, from the body of chickens and turkeys in Europe and North America. The latter species is about 0.4 mm. long. Alicata *et al.* (1946) were able to reduce drastically the numbers of body mites, *Megninia cubitalis*, by applying 5 per cent DDT, or undiluted sodium fluoride, or undiluted sodium fluosilicate to Hawaiian chickens. They also found that good control could be obtained for the wing mite, *Pterolichus obtusus*, by using 10 per cent "Lethane A-70" or by undiluted "NH Dust."

Levi (1941) states that pigeons in South Carolina may have the feathers of the neck and body infested by *M. columbae*. Reis (1939), of Brazil, also reports this mite in pigeons and adds another similar species, *M. ginglymura* of chickens.

*Sternostomum rhinolethrum* is a stout-bodied, rather heavy-legged mite related to the common red mite (*Dermanyssus gallinae*). It is said to occur in the nasal fossae of chickens, sucking blood and causing catarrhal rhinitis. The adult mite is about 1 mm. in length, and the legs are armed with recurved hooklets for attachment to the host. Similar species have been reported from ducks and young pigeons.



FIG. 31.20. *Falculifer rostratus.* A feather mite of pigeons, male. ×68. (Reis and Nobrega.)

In addition to the mites listed, numerous other species have been noted on birds in various parts of the world. Some of these no doubt will be found invading domestic poultry. For the present they may be considered of minor importance.

## TICKS OF POULTRY

Although ticks are quite important parasites of mammals, they are of relatively minor significance to birds.

Both ticks and mites are invertebrate arthropods of the class **ARACH-NIDA**, order ACARINA. The ticks constitute a blood-sucking superfamily, namely the Ixodoidea. They are distinguished from the mites by being usually larger (up to 15 mm. in length) and by the presence of a pair of respiratory openings, one on each side of the leathery abdomen. These openings, called spiracles or stigmal plates are situated either between the bases of the last two pairs of legs (family Argasidae) or posterior to the last pair (family Ixodidae).

A typical tick life cycle includes the egg, the larva (seed tick), the nymph, and the adult stages. After engorging with blood, the female tick drops from the host to hide in soil, humus, litter, tree bark, or crevices during the pre-ovulation and egg-laying periods. From several hundred to several thousand eggs are laid. This takes weeks to months after which the female tick dies. The male previously had died following copulation. After an incubation period, the minute six-legged larvae (seed ticks) emerge from the egg shells and await contact with a suitable host. Feeding on blood is followed by molting of the larval skin and emergence of the eight-legged nymphs that resemble the adults except for maturity of the reproductive organs. One or more additional molts follow before the adult female and male ticks fully develop.

The total length of the tick life cycle varies greatly, depending upon the species of tick, availability of suitable hosts, and climatic conditions. Some ticks complete the cycle within six weeks; others may require two years. In general, warmth and relative dryness favor tick development, although many species can withstand extremely cold weather. On vacated premises the adults especially may remain alive for many months or even several years.

Disease caused by ticks may be of three general types. Foremost is the loss of host blood, which may result fatally. Secondly, there must be considered loss in production, no doubt associated with blood loss, but also possibly due to tick-produced toxic substances. Thirdly, ticks in general are notorious transmitters of other parasites, such as those of avian spirochetosis, tularemia, babesiosis, anaplasmosis, dirofilariasis, encephalomyelitis, and certain rickettsial diseases, notably Rocky Mountain spotted fever. The first two diseases are of particular interest to poultry raisers. Other avian diseases may be associated with tick transmission as has been suggested by Brown and Cross (1941) for avian leukosis.

Few species of ticks are host-specific, and those found on birds are no exceptions. The principal ticks reported from birds in North America are:

*Argas persicus,* the chicken tick that also occurs on geese, ducks, turkeys, guinea fowl, ostriches, pigeons, canaries, various wild birds, and rarely on cattle and on man (see page 751).

*Argas reflexus,* the pigeon tick, has been found on ducks, geese, English

sparrows, and other wild birds; also on monkeys, rabbits, horses, and man (see page 753).

*Otobius megnini,* the spinose ear tick of mammals such as horses, cattle, sheep, goats, dogs, cats, and man, may also occur on ostriches. The second nymphal stage is that usually seen as a parasite.

*Ornithodoros coriaceus,* the Pajaroello tick, a venomous species, has been reported from various mammals and from various birds.

*Ornithodoros turicata* may infest various birds, reptiles, and mammals.

*Ixodes brunneus* has been reported from various bird hosts.

*Rhipicephalus sanguineus,* the brown dog tick, has also been collected from two species of wild birds, although it usually infests dogs, cats, horses, cattle, sheep, man, and various wild mammal species.

*Haemaphysalis leporis-palustris,* the rabbit tick, is found mainly on rabbits and hares, also on dogs, cats, horses, and rarely on man. It may infest quail and various other birds.

*Haemaphysalis cinnabarina* has been reported from various bird hosts including the turkey. It also occurs on mammals.

*Haemaphysalis chordeilis* has been taken from various species of game birds.

*Amblyomma maculatum,* the Gulf coast tick, parasitizes wild birds, also wild and domestic mammals.

*Argas persicus,* the fowl tick (Fig. 31.21), is the most important tick parasite of birds. Among its many common names are chicken tick, blue "bug," tampan, and adobe tick. In the United States it is distributed mainly in those states along the Gulf of Mexico and the Mexican border. It is also established in many other tropical and temperate areas of the world.

The mature, blood-engorged female measures about 10 mm. in length and about 6 mm. in width, the mature male being about half that size. This tick is relatively easily recognized by its flattened ovoid shape and reddish-brown color. There is no scutum or dorsal shield, which thus distinguishes it as belonging to the tick family Argasidae. The mouthparts are on the ventral anterior surface, hidden from above by the projecting body.

The female fowl tick lays up to a total of about 700 eggs at several layings, between which she seeks a host for a meal of blood. The eggs are laid in sheltered crevices, including the bark of trees. They hatch in from 10 days in warm weather to three months during cool periods. The almost microscopic larvae or seed ticks immediately seek a host, although they may live for several months without eating. After feeding on blood for several days, the larvae leave the host for a hiding place nearby, and in from about 4 to 9 days they reach the nymphal stage. Nymphs may do without food for as long as fifteen months. However, if a host is available, the nymphs feed during a night,

then hide for 10 or 12 days, shed their skins, and reach a second nymphal stage. After about an hour's feeding at night and about a week in hiding, the adult ticks emerge from the nymphal skins, now ready to engorge with blood and reproduce over a period of about 30 days. The adult fowl tick may live for more than two years in an unfed state.

Birds suffer to the greatest degree from attacks of these ticks during the warmer, dryer seasons of the year. The loss of blood may reach the proportions of a fatal anemia. At least there may be expected to be emaciation,
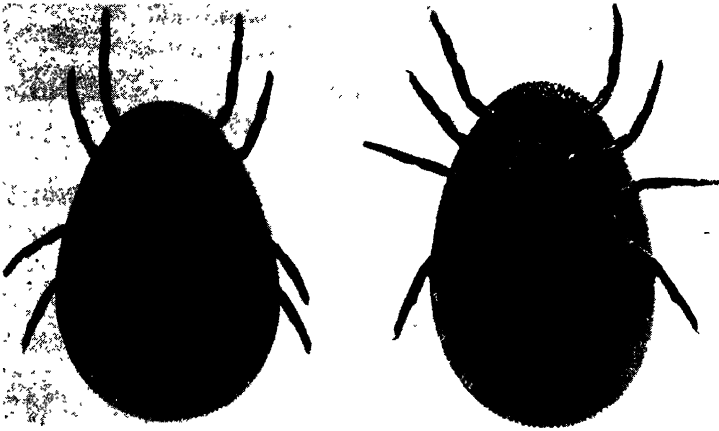


Fig. 31.21. *Argas persicus.* The fowl tick or blue "bug," female, nearly engorged with blood. Dorsal and ventral views. (U.S.D.A., Bur. Entomol. and Plant Quar.)

weakness, slow growth, and lowered production. Ruffled feathers, poor appetite, and diarrhea are symptoms suggesting tick infestation. Turkeys usually suffer even more than chickens, and recently hatched poults and chicks show the highest mortality rate.

The fowl tick is capable of transmitting a spirochete from the blood of infected birds to that of susceptible birds in many parts of the world. This organism, *Treponema anserinum (Borrelia gallinarum)* is highly pathogenic, but thus far it has not been reported from the United States. Apparently Brazil is the area nearest to the United States in which avian spirochetosis occurs. Mönnig (1938) states that *Argas persicus* may also act as a vector of *Aegyptianella pullorum,* an erythrocyte-invading protozoan (piroplasm) of birds. The disease thus produced, aegyptianellosis, has not been reported from the Americas. Brown and Cross (1941) found evidence that fowl ticks are agents for the transmission of the virus of fowl paralysis. Howell, Stiles, and Moe (1943) reported that occasionally the fowl tick may transmit the protozoan parasite of anaplasmosis of cattle.

The control of the fowl tick is carried out in a manner similar to that used for the control of the common red mite, *Dermanyssus gallinae* (page 738). Mönnig (1938) recommends that the birds be placed in wooden crates away from the houses or rubbish piles. Then the chicken house should be cleaned and sprayed. After about 10 days, all larval ticks will have dropped from the birds, which may then be returned to the cleaned house. The crates are made tick-free by scalding or spraying, or they may be burned.

Bishopp (1941) recommends a control program, starting in the morning, with the removal of birds from the house. Then all litter and useless material is removed from the house and sprayed or burned. The house itself is thoroughly disinfested by spraying, using one of the carbolineums or crude oil 2 parts thinned with kerosene 1 part. The birds are not returned to the house until the spray has dried. Coops and brooders should be similarly made tick-free. The foregoing operation should be repeated one or more times at 30-day intervals, although the spray material may be applied with a brush if the first application has been done with a spray pump. Bishopp (1941) also describes the construction of sanitary chicken roosts, set in the floor away from the sidewalls so that the birds do not contact the walls while roosting. Simply constructed, easily demountable nesting boxes should also be used.

Dove (1945) quotes Parish as having successfully treated houses infested with the fowl tick by using 5 per cent DDT in kerosene as a spray, forcing the spray into cracks and crevices. For 3 months following treatment no living ticks could be found. On account of the long life of this tick, the test will be observed for any reoccurrence of the infestation.

Other methods for the control of fowl ticks include the use of metal construction, the elimination of tree roosting, using roosts suspended from the ceiling, and sulfur fumigation of tightly constructed houses. Frequent inspection is necessary in order to combat ticks before their number has increased to a harmful extent.

*Argas reflexus*, the pigeon tick, probably occurs to some extent in this country. It is quite similar to the fowl tick, *Argas miniatus*, in appearance and habits. The sides of the body narrow considerably toward the front. Control measures are essentially the same as for the fowl tick.

### REFERENCES

Abbott. W. S.: 1920. Results of experiments with miscellaneous substances against chicken lice and the dog flea. U.S.D.A., Bul. 888.

Alicata, J. E.: 1942. Experimental transmission of endemic typhus fever by the sticktight flea. *Echidnophaga gallinacea.* Jour. Wash. Acad. Sci. 32:57.

——, Holdaway, F. G., Quisenberry, J. H., and Jensen, D. D.: 1946. Observations on the comparative efficacy of certain old and new insecticides in the control of lice and mites of chickens. Poultry Sci. 25:376.

André, M.: 1930. Contribution à l'étude d'un acarien: le *Trombicula autumnalis* Shaw. Mémoirs de la Soc. Zool. de France 29:39.

Anonymous: 1945. A new British insecticide (Hexachlorocyclohexane or 666). Soap and San. Chem. 21 (5) :103.

Annand, P. N., and Members of Staff: 1944. Tests conducted by the Bureau of Entomology and Plant Quarantine to appraise the usefulness of DDT as an insecticide. Jour. Econ. Entomol. 37:125.

Back, E. A., and Bishopp, F. C.: 1942. Bedbugs as pests of poultry. U.S.D.A., Yearbook, p. 1068.

———— and Cotton, R. T.: 1938 (revised). Stored grain pests. U.S.D.A., Farmers' Bul. 1260. (Beetles and beetle larvae.)

Baker, A. D.: 1933. Some studies of the dipterous fauna of the poultry yard in Quebec in relation to parasitic troubles. Poultry Sci. 12:42.

Banks, N.: 1915. The Acarina or mites. U.S.D.A., Office of Secretary, Rep. 108.

Bare, O. S.: 1943. External parasites of poultry and methods for their control. Nebr. Agr. Exper. Sta., Cir. 75.

Barger, E. H., and Card, L. E.: 1938. Diseases and Parasites of Poultry. 2nd ed. Lea and Febiger, Philadelphia.

Barnes, S.: 1945. The residual toxicity of DDT to bedbugs (Cimex lectularius). Lond. Sch. of Trop. Hyg., Ministry of Prod., Insecticides Development Panel, Brit. Apt. IDP (45) :226.

Bedford, G. A. H.: 1924. The external parasites of poultry, with measures for their control. Jour. Union So. Africa Dept. Agr. 9:123.

Beller, K. F.: 1924. Dermanyssus avium als Ursache von Massensterben beim Hausgeflügel. Berliner tierärztl. Wochenschr. 40:261.

Bequaert, J.: 1925. Notes on Hippoboscidae. Psyche 32:265.

————: 1935. Notes on Hippoboscidae. 9. A further study of Pseudolynchia. Rev. Zool. Bot. Africa 27:395.

Bishopp, F. C.: 1922. Fleas and their control. U.S.D.A., Farmers' Bul. 897.

————: 1923. Limberneck of fowls produced by fly larvae. Jour. Parasit. 9:170.

————: 1929. The pigeon fly, an important pest of pigeons in the United States. Jour. Econ. Entomol. 22:974.

————: 1933. Mosquitoes kill livestock. Science 77:115.

————: 1941. The fowl tick. U.S.D.A., Farmers' Bul. 1070 (revised).

————: 1942a. Poultry lice and their control. U.S.D.A., Yearbook, p. 1048.

————: 1942b. The pigeon fly. U.S.D.A., Yearbook, p. 1072.

———— and Wagner, R. D.: 1931. Nicotine in the control of ectoparasites of poultry. Jour. Econ. Entomol. 24:56.

———— and Wood, H. P.: 1931. Mites and lice on poultry. U.S.D.A., Farmers' Bul. 801.

Blacklock, B.: 1912. On the resistance of Cimex lectularius to various reagents, powders, liquids, and gases. Annals Trop. Med. and Parasit. 6:415.

Brody, A. L.: 1936. The transmission of fowl pox. (By mosquitoes.) Cornell Univ. Agr. Exper. Sta., Memoir 195.

Brown, J. C., and Cross, J. C.: 1941. A probable agent for the transmission of fowl paralysis. (Fowl tick.) Science 93:528.

Bunyea, H., and Wehr, E. E.: 1941. Diseases and parasites of poultry. U.S.D.A., Farmers' Bul. 1652:73.

Bureau of Entomology and Plant Quarantine, U.S.D.A.: 1946. DDT and other insecticides and repellents developed for the armed forces. U.S.D.A., Misc. Pub. 606.

Bushnell, L. D., and Twiehaus, M. J.: 1939. Poultry diseases, their prevention and control. Kan. Agr. Exper. Sta., Bul. 284 (revised) :83.

Cameron, D.: 1938. The northern fowl mite (Liponyssus sylviarum). Investigations at Macdonald College, Quebec, with a summary of previous work. Canad. Jour. Res. 16:230 (Sec. D).

Carpenter, C. D.: 1931. The use of nicotine and its compounds for the control of poultry parasites. Jour. Am. Vet. Med. Assn. 78:651.

Chandler, W. L.: 1930. (Orthophenylphenol as a parasiticide.) Mich. St. Bd. Agr., Ann. Rep. 69:184.

————: 1931. (Orthophenylphenol as a delousing agent.) Mich. St. Bd. Agr., Ann. Rep. 70:236.

Chapin, E. A.: 1925. Freyana (Microspalax) chaneyi from a turkey, Meleagris gallopavo. Jour. Parasit. 12:113.

Coatney, G. R.: 1931. On the biology of the pigeon fly, Pseudolynchia maura Bigot. Parasitology 23:525.

Cooley, R. A., and Kohls, G. M.: 1944. The Argasidae of North America, Central America, and Cuba. The Univ. Press, Notre Dame, Ind.

Cotton, R. T., and St. George, R. A.: 1929. The meal worms. (Beetles and beetle larvae.) U.S.D.A., Tech. Bul. 95.

Creel, R. H., and Faget, F. M.: 1916. Cyanide gas for the destruction of insects. U. S. Pub. Health Service, Reprint 343 from Pub. Health Reps., p. 1464.

Creighton, J. T., Dekle, G. W., and Russell, J.: 1943. The use of sulfur and sulfur compounds in the control of poultry lice. Jour. Econ. Entomol. 36:413.

Crutchfield, C. M., and Hixson, H.: 1943. Food habits of several species of poultry lice, with special reference to blood consumption. Fla. Entomol. 26:63.

Cutright, C. R.: 1929. A valuable aid in the control of the feather mite, *Liponyssus sylviarum*. Jour. Econ. Entomol. 22:422.

Davidson, W. M.: 1924. Results of experiments with miscellaneous substances against the chicken mite. U.S.D.A., Dept. Bul. 1228.

Davis, W. A.: 1940. A study of birds and mosquitoes as hosts for the virus of eastern equine encephalomyelitis. Am. Jour. Hyg., Sec. C, 32:45.

Deakin, A., and Robertson, G.: 1933. Effect of mercurial ointment on hatchability. Poultry Sci. 12:378.

de Oliveira Castro, G. M.: 1930. The transmission of epithelioma contagiosa by mosquitoes (trans. title). Compt. rend. Soc. de biol. (Paris) 105:316.

de Zayas, F.: 1941. Los malphagos de las aves domesticas en Cuba. Univ. Havana, Mem. de la Soc. Cub. de Hist. Nat. 15:201.

Dietrich, A.: 1925. *Laminosioptes cysticola* und *Cytoleichus sarcoptoides* bei Hühnern. Berliner tierärztl. Wochenschr. 41:486.

Dove, W. E.: 1945. Summary of DDT experiments on insects that affect man and animals. U.S.D.A., Bur. Entomol. and Plant Quar., Mimeo. Cir. E-673.

Drake, C. J., and Jones, R. M.: 1930. The pigeon fly and pigeon malaria in Iowa. Ia. St. Coll. Jour. Sci. 4:253.

Emmel, M. W.: 1937. Sulfur in the control of external parasites of chickens. Preliminary report. Jour. Am. Vet. Med. Assn. 91:201.

————: 1942. Field experiments in the use of sulfur to control lice, fleas, and mites of chickens. Fla. Agr. Exper. Sta., Bul. 374.

Ewing, H. E.: 1911. The English sparrow as an agent in the dissemination of chicken and bird mites. Auk. 28:335.

————: 1921. Studies on the biology and control of chiggers. U.S.D.A., Bul. 986.

————: 1923. The dermanyssid mites of North America. Proc. U. S. Nat. Museum, 62, Art. 13:1026.

————: 1929a. A Manual of External Parasites. C. C. Thomas, Springfield, Illinois.

————: 1929b. Birds as hosts for the common chigger. Amer. Nat. 63:94.

————: 1938. A key to the genera of chiggers (mite larvae of the subfamily Trombiculinae) with descriptions of new genera and species. Jour. Wash. Acad. Sci. 28:288.

————: 1944. The trombiculid mites (chigger mites) and their relation to disease. Jour. Parasit. 30:339.

———— and Fox, I.: 1943. The fleas of North America. U.S.D.A., Misc. Pub. 500.

Fox, Irving: 1940. Fleas of Eastern United States. Iowa State College Press, Ames, Iowa.

Gallagher, B. A.: 1920. Rose-chafer poisoning in chickens. Jour. Am. Vet. Med. Assn. 57:692.

Gelormini, N., and Roveda, R. J.: 1941. Sarna epidermóptica en el canario. Univ. Buenos Aires, Rev. Fac. Agron. y Vet. 9:143.

Gibson, A.: 1930. Insect and other external parasites of poultry in Canada. Sci. Agr. 11:208.

Graesser, F. E.: 1943. Scabies in a turkey. Canad. Jour. Comp. Med. 7:13.

Guberlet, J. E., and Hotson, H. H.: 1940. A fly maggot attacking young birds, with observations on its life history. Murrelet 21:65.

Hall, M. C.: 1929. Arthropods as intermediate hosts of helminths. Smithsonian Inst., Misc. Collections 81:1–77.

Hart, L.: 1938. A short note on the transmission of the fowl spirochaete (*Treponema anserinum*) by red mite (*Dermanyssus gallinae*). Vet. Res. Rep., Dept. Agr., N. S. Wales, No. 7:74.

Hearle, E.: 1938. Insects and allied parasites injurious to livestock and poultry in Canada. Canad. Dept. Agr., Publ. 604.

Herman, C. M.: 1938a. Mosquito transmission of avian malaria parasites. Am. Jour. Hyg., Sec. C, 27:345.

Herman, C. M.: 1938b. Occurrence of larval and nymphal stages of the rabbit tick, *Haemaphysalis leporis–palustris*, on wild birds from Cape Cod. Bul. Brooklyn Entomol. Soc. 33:133.

Herms, W. B.: 1939. Medical Entomology. 3rd ed. The Macmillan Co., New York.

Herrick, G. W.: 1915. Some external parasites of poultry with special reference to Mallophaga, with directions for their control. Cornell Univ. Agr. Exper. Sta., Bul. 359.

Hertel, M.: 1904. Über Geflügelcholera und Hühnerpest. Arb. a.d. kaiserl. Gesundheitsamt. 20:453.

Hinshaw, W. R.: 1937. Diseases of turkeys. Calif. Agr. Exper. Sta., Bul. 613:108.

Hipolito, O., and [de] Freitas, M. G.: 1943. Notas ornitopatologicas. Observacoes sôbre alguns acarinos parasitos de *Gallus gallus domesticus*, em Minas. Arq. Escola Superior Vet., Estado Minas Gerais (Brazil) 1:81.

Hirst, S.: 1922. Mites injurious to domestic animals. Brit. Museum, Econ. Series 13:1–107.

Houbrich, W., and Gilbert, T. J.: 1922. Blood sucking flies. Am. Pigeon Jour. 11:383.

Howard, L. O., and Popenoe, C. H.: 1916. Hydrocyanic-acid gas against household insects. U.S.D.A., Farmers' Bul. 699.

Howell, D. E., Stiles, G. W., and Moe, L. H.: 1943. The fowl tick (*Argas persicus*), a new vector of anaplasmosis. Am. Jour. Vet. Res. 4:73.

Hoyle, W. L.: 1938. Transmission of poultry parasites by birds, with special reference to the "English" or house sparrow and chickens. Trans. Kan. Acad. Sci. 41:379.

Huff, C. G.: 1932. Further infectivity experiments with mosquitoes and bird malaria. Am. Jour. Hyg. 15:751.

Hungerford, T. G.: 1938. Field observations on spirochaetosis (*Spirochaeta anserina*) of poultry, transmitted by the red mite (*Dermanyssus avium*) in New South Wales. Vet. Research Rep., Dept. Agr., N.S.W., No. 7:71.

Illingworth, J. F.: 1915. Notes on the habits and control of the chicken flea (*Echidnophaga gallinacea* Westwood). Jour. Econ. Entomol. 8:492.

James, W. A., Graham, R., and Thorp, F.: 1930. Epidermoptic scabies in a hen. Jour. Am. Vet. Med. Assn. 76:93.

Johnson, E. P., Underhill, G. W., Cox, J. A., and Threlkeld, W. L.: 1938. A blood protozoan of turkeys transmitted by *Simulium nigroparvum* (Twinn). Am. Jour. Hyg. 27:649.

Jordan, K.: 1929. Further records of North American bird-fleas, with a list of the Nearctic birds from which fleas are known. Novit. Zool. 35:89.

Kadner, C. G.: 1941. Pigeon malaria in California. Science 93:281.

Kasparek, A.: 1907. Bericht über die 79. Versammlung Deutscher Naturforscher und Aerzte in Dresden. Deutsch. tierärztl. Wochenschr. 15:623. (*Laminosioptes cysticola*.)

Kéler, S.: 1938. Übersicht über die gesamte Literatur der Mallophagen (1668–1938). Zeitschr. angew. Entomol. 25:487.

Kellogg, V. L.: 1900. A list of the biting lice (Mallophaga) taken from birds and mammals of North America. Proc. U. S. Nat. Museum 22:39.

Kitselman, C. H., and Grundmann, A. W.: 1940. Equine encephalomyelitis virus isolated from naturally infected *Triatoma sanguisuga* Le Conte. Kan. Agr. Exper. Sta., Tech. Bul. 50.

Kligler, I. J., Muckenfuss, R. S., and Rivers, T. M.: 1929. Transmission of fowl pox by mosquitoes. Jour. Exper. Med. 49:649.

Knipling, E. F., and Rainwater, H. T.: 1937. Species and incidence of dipterous larvae concerned in wound myiasis. Jour. Parasit. 23:451.

Köhnlein, J.: 1925. Die Vogelmilbe (*Dermanyssus avium*) und ihre Bekämpfung. Arch. f. wissen. Tierheilk. 53:144.

Kulash, W. M., and Maxwell, J. M.: 1945. DDT and bedbugs in chicken houses. Jour. Econ. Entomol. 38:606.

Lahaye, J.: 1930. Traitement des maladies parasitaires des volailles. Rep. Internat. Vet. Cong., London, p. 801.

Lamson, Jr., G. H.: 1917. Mercurial ointment, an effective control of hen lice. Jour. Econ. Entomol. 10:71.

————: 1922. The rose chafer as a cause of death of chickens. Storrs Agr. Exper. Sta., Bul. 110:117.

Lesbouyries, G.: 1941. La Pathologie des Oiseaux. Vigot Frères, Paris. p. 816.

Levi, W. M.: 1941. The Pigeon. R. L. Bryan Co., Columbia, S. C. p. 314.

Madden, A. H., Lindquist, A. W., and Knipling, E. F.: 1944. Tests of repellents against chiggers. Jour. Econ. Entomol. 37:283.

————, Lindquist, A. W., and Knipling, E. F.: 1945. DDT and other insecticides as residual-type treatments to kill bedbugs. Jour. Econ. Entomol. 38:265.

Marcovitch, S.: 1926. The control of poultry lice and mites. (Sodium fluosilicate.) Tenn. Agr. Exper. Sta., Cir. 2.

Marlatt, C. L.: 1925. The bedbug. U.S.D.A., Farmers' Bul. 754.

Martin, M.: 1934. Life history and habits of the pigeon louse *Columbicola columbae* (Linn.). Canad. Entomol. 66:6.

Matheson, R., Brunett, E. L., and Brody, A. L.: 1931. The transmission of fowl pox by mosquitoes. Preliminary report. Poultry Sci. 10:211.

Maw, W. A., Whitehead, W. E., and Bemont, L. H.: 1935. The northern fowl mite and its control. Sci. Agr. 16:79.

Mégnin, P.: 1879. Les acariens parasites du tissue cellulaire et des réservoirs aériens chez les oiseaux. Jour. Anat. et Physiol. 15:123.

Melvin, R., Smith, C. L., Parish, H. E., and Barrett, Jr., W. L.: 1941. A new remedy for the prevention and treatment of screwworm infestation of livestock. U.S.D.A., Bur. Entomol. and Plant Quar., Pub. E-540 (mimeographed).

Metz, K.: 1911. *Argas reflexus*, die Taubenzecke. Monatschr. f. prakt. Tierheilk. 22:481.

Mönnig, H. O.: 1938. Veterinary Helminthology and Entomology. 2nd ed. William Wood and Co., Baltimore, Maryland.

Myers, L. E.: 1928. The American swallow bug. *Oeciacus vicarius* Horváth. Parasit. 20·159.

Neumann, L. G.: 1909. Parasites et Maladies Parasitaires des Oiseaux Domestiques. Asselin et Houzeau, Paris.

Neveu-Lemaire, M.: 1938. Traité d'Entomologie Médicale et Vétérinaire. Vigot Frères, Paris.

Nuttall, G. H. F., and Warburton, C.: 1908. Ticks, a Monograph of the Ixodoidea. Part I, Argasidae; Part II, Ixodidae. Cambridge (England) Univ. Press.

Olson, C.: 1935. The effect of certain ectoparasites on the cellular elements and hemoglobin of the blood of the domestic chicken. Jour. Am. Vet. Med. Assn. 87:559.

O'Roke, E. C.: 1930. The morphology, transmission, and life history of *Haemoproteus lophortyx* O'Roke, a blood parasite of the California valley quail. Univ. of Calif. Pub. in Zool. 36:1–50

————: 1934. A malaria-like disease of ducks, caused by *Leucocytozoon anatis* Wickware. Univ. of Mich. School of Forestry and Conserv., Bul. 4.

Osborn, H.: 1896. Insects affecting domestic animals. U.S.D.A., Div. Entomol., Bul. 5 n.s.

Parish, H. E.: 1942. Phenothiazine protects chickens from lice in Texas test. E. I. du Pont de Nemours and Co., Agr. News Letter 10:35.

————: 1943. Sodium fluosilicate to control poultry lice. Jour. Econ. Entomol. 36:353.

Parman, D. C.: 1923. Biological notes on the hen flea, *Echidnophaga gallinacea*. Jour. Agr. Res. 23:1007.

————, Abbott, W. S., Culver, J. J., and Davidson, W. M.: 1928. Ineffectiveness of internal medication of poultry for the control of external parasites. U.S.D.A., Tech. Bul. 60.

Patton, J. W.: 1921. The fowl tick, *Argas miniatus*. Poultry Sci. 1:125.

Payne, L. F.: 1929. A new method of controlling feather mites. Jour. Econ. Entomol. 22:819.

————: 1930. Feather mites and their control. Ala. Polytech. Inst., Bul. Vol. 25 (1) pp. 61–63 (1930). Twenty-first Ann. Meeting, Proc. of Poultry Sci. Assn.

Peters, H. S.: 1936. A list of external parasites from birds of the eastern part of the United States. Bird Banding 7:9.

Pillers, A. W. N.: 1921. Notes on mange, and allied mites for veterinarians. Baillière, Tindall, and Cox, London.

————: 1927. Perforations in pigeons' feathers due to the mite, *Falculifer rostratus* (Buchholz). Vet. Jour. 83:410.

Pinto, C.: 1930. Arthropodos Parasitos e Transmissores de Doencas. Mello e C., Rio de Janeiro.

Plasaj: 1925. Sur la transmission du choléra aviaire par les *Dermanyssus*. Jugoslav. Vet. Glasnik., Livr. 1–6. (Rev. in Rev. Gén. de Méd. Vét. 34:654.)

Povar, M L.: 1946. Value of DDT for the control of the northern feather mite, *Liponyssus sylviarum*. Cornell Vet. 36:91.

Prouty, M. J., and Coatney, G. R.: 1934. Further studies on the biology of the pigeon fly, *Pseudolynchia maura* Bigot. Parasit. 26:249.

Quigley, G. D., and Cory, E. N.: 1946. The utility of DDT for the control of poultry ectoparasites. Poultry Sci. 25:419.

Railliet, A.: 1895. Traité de Zoologie Médicale et Agricole. 2e édition. Asselin et Houzeau, Paris.

Readio, P. A.: 1927. Studies on the biology of the Reduviidae of America north of Mexico. Univ. of Kan., Sci. Bul. 17:5.

Rebrassier, R. E., and Martin, E. D.: 1932. *Syringophilus bipectinatus,* a quill mite of poultry. Science 76:128.

Reeves, W. C., Hammon, W. M., Furman, D. P., McClure, H. E., and Brookman, B.: 1947. Recovery of western equine encephalomyelitis virus from wild bird mites (*Liponyssus sylviarum*) in Kern County, California. Science 105:411.

Reid, W. M., and Ackert, J. E.: 1937. The cysticercoid of *Choanotaenia infundibulum* (Bloch) and the housefly as its host. Trans. Amer. Micro. Soc. 56:99.

Reis, J.: 1939. Alguns parasitas de *Gallus gallus* (L.) verificados em São Paulo. Arq. Inst. Biol. 10:147

—— and Nobrega, P.: 1936. Tratado de Doencas das Aves. Instituto Biologico, São Paulo, Brazil.

Riley, W. A, and Johannsen, O. A.: 1938. Medical Entomology. McGraw-Hill Book Co., New York.

Sanborn, C. E.: 1919. The chicken sticktight flea. Okla. Agr. Exper. Sta., Bul. 123.

Schalk, A. F.: 1928. Results of some avian tuberculosis studies. (Transmission by fly larvae.) Jour. Am. Vet. Med. Assn. 72:852.

Scott, E. W., Abbott, W. S., and Dudley, J. E.: 1918. Results of experiments with miscellaneous substances against bedbugs, cockroaches, clothes moths, and carpet beetles. U.S.D.A., Bul. 707.

Shew, W. D.: 1944. Livestock notes. Lice on fowls. Jour. Dept. Agr., Victoria 42:215.

Shillinger, J. E.: 1937. Diseases of upland game birds. U.S.D.A., Farmers' Bul. 1781.

Skidmore, L. V.: 1932a. *Leucocytozoon smithi* infection in turkeys and its transmission by *Simulium occidentale* Townsend. Zentralbl. f. Bakt. I. Orig. 125:329.

——: 1932b. The transmission of fowl cholera to turkeys by the common house fly (*Musca domestica* Linn.) (with brief notes on the viability of fowl cholera microorganisms). Cornell Vet. 22:281.

Smith, C. N., and Gouck, H. K.: 1944. DDT, sulfur and other insecticides for the control of chiggers. Jour. Econ. Entomol. 37:131.

Smith, M. G., Blattner, R. J., and Heys, F. M.: 1944. The isolation of the St. Louis encephalitis virus from chicken mites (*Dermanyssus gallinae*) in nature. Science 100:362.

Snipes, B. T., Carvalho, J. C. M., and Tauber, O. E.: 1940. Biological studies of *Ornithocoris toledoi* Pinto, the Brazilian chicken bedbug. Ia. St. Coll. Jour. Sci. 15:27.

Stage, H. H.: 1946. The use of DDT in controlling fleas. U.S.D.A., Bur. Entomol. and Plant Quar., Report E-680.

Steward, J. S.: 1947. Application of "gammexane" to arthropods of veterinary importance. Vet. Record 59:27.

Stewart, M. A.: 1927. A means of control of the European hen flea, *Ceratophyllus gallinae* Schrank. Jour. Econ. Entomol. 20:132.

——: 1929. A case of cloacal myiasis in a hen and its treatment. Cornell Vet. 19:49.

——: 1932. Dispersal of the sticktight flea of hens (*Echidnophaga gallinacea* Westw.). Jour. Econ. Entomol. 25:164.

Stoddard, H. L.: 1932. The Bobwhite Quail, Its Habits, Preservation and Increase. Chas. Scribner's Sons, New York.

Sulkin, S. E.: 1945. Recovery of equine encephalomyelitis virus (western type) from chicken mites. Science 101:381.

Sullivan, K. C.: 1924. The use of calcium cyanide for the control of fleas and other insects. Jour. Econ. Entomol. 17:230.

Telford, H. S.: 1945a. New insecticides for chicken lice control. Jour. Econ. Entomol. 38:573.

——: 1945b. DDT as a chicken louse control. Jour. Econ. Entomol. 38:700.

Underhill, G. W.: 1939. Two simuliids found feeding on turkeys in Virginia. Jour. Econ. Entomol. 32:765.

——: 1944. Blackflies found feeding on turkeys in Virginia (*Simulium nigroparvum* Twinn and *S. slossonae* Dyar and Shannon). Va. Agr. Exper. Sta., Tech. Bul. 94.

Van Es, L., and Olney, J. F.: 1941. Poultry diseases and parasites. Nebr. Agr. Exper. Sta., Bul. 332.

van Heelsbergen, T.: 1929. Handbuch der Geflügelkrankheiten und der Geflügelzucht. F. Enke, Stuttgart.

Wakeland, C.: 1935. Fumigation for the control of household insects. (Bedbugs.) Univ. Ida. Agr. Ext. Circ. 50.

Walker, G. P.: 1927. A blackfly (*Simulium bracteatum*) fatal to goslings. Canad. Entomol. 59:123.

Ward, A. R., and Gallagher, B. A.: 1920. Diseases of Domesticated Birds. The Macmillan Co., New York.

Warren, D. C.: 1945. The value of DDT for the control of the common chicken louse. Poultry Sci. 24:473.

Wells, R. W., Bishopp, F. C., and Laake, E. W.: 1922. Derris as a promising insecticide. Jour. Econ. Entomol. 15:90.

Whitehead, W. E.: 1942. Lice and some other external parasites of domestic animals and poultry in the province of Quebec. Macdonald College, McGill Univ., Farm Bul. 7.

————— and Maw, W. A.: 1934. Control of the northern fowl mite. Sci. Agr. 15:126.

Wickware, A. B.: 1921. An unusual form of scabies in fowls. Jour. Parasit. 8:90. (*Megninia gallinulae.*)

Wilkins, S. D., and Dutcher, R. A.: 1920. Limberneck in poultry. (Fly larvae.) Jour. Am. Vet. Med. Assn. 57:653.

Wilson, F. H.: 1933. A louse feeding on the blood of its host. Science 77:490. (*Menopon stramineum.*)

—————: 1941. The slender lice of American pigeons and doves, with descriptions of two new species. Jour. Parasit. 27:259.

Wisseman, Jr., C. L., and Sulkin, S. E.: 1945. Life cycle and care of the chicken mite (*Dermanyssus gallinae*) in the laboratory. (An abstract.) Jour. Bact. 50:128.

Wolfenbarger, D. O., and Hoffmann, E.: 1944. Uses of DDT on the poultry farm. Poultry Sci. 23:545.

Wolffhügel, K.: 1910. Die Flöhe (Siphonaptera) der Haustiere. Zeitschr. f. Infekt-Krankh. d. Haustiere 8:218 and 354.

Wood, H. P.: 1917. The chicken mite, its life history and habits. U.S.D.A., Bul. 553.

—————: 1919. The depluming mite of chickens: its complete eradication from a flock by one treatment. Jour. Econ. Entomol. 12:402. (*Cnemidocoptes gallinae.*)

—————: 1920. Tropical fowl mite in the United States. U S.D.A., Dept. Circ. 79.

—————: 1922. Eradication of lice on pigeons. U.S.D.A., Dept. Circ. 213.

# NEMATODES AND ACANTHOCEPHALIDS OF POULTRY

By Everett E. Wehr, *Zoological Division, Bureau of Animal Industry, United States Department of Agriculture, Washington, D. C.*

☆ ☆ ☆

## NEMATODES

Nematodes or roundworms are usually elongated, cylindrical, and unsegmented worms. The body is covered with a tough, noncellular layer known as the cuticle. These worms have a well-developed alimentary tract and, in contrast to the tapeworms, are usually bisexual.

According to Chitwood and Chitwood (1937), the class **NEMATODA** is divided into two subclasses, PHASMIDIA and APHASMIDIA. The subclass PHASMIDIA is characterized by the presence of phasmids and porelike amphids, and the absence of caudal glands. This subclass is subdivided into two orders; namely, Rhabditida and Spirurida. The order Rhabditida is composed of many free-living nematodes and a few parasitic forms, while the order Spirurida contains purely parasitic forms. Of the thirty families comprising the order Rhabditida, only five families (Strongyloididae, Syngamidae, Heterakidae, Ascarididae, and Trichostrongylidae) contain parasitic nematodes commonly found in poultry. The order Spirurida includes thirteen families, three of which (Thelaziidae, Spiruridae, and Acuariidae) only contain forms parasitizing poultry.

The subclass APHASMIDIA is characterized by having amphids which are spiral, circular, vesiculate, tuboid, or rarely porelike and by the absence of phasmids. The members of this group are divided into two orders; namely, Chromadorida and Enoplida. These two orders are distinguished by the spiral, circular, and vesiculate amphids and by the esophagus being divisible into three regions in the former, and the pocket-like or porelike amphids and by the esophagus being divisible into only two parts in the latter. The order Chromadorida does not include any species of nematodes occurring as parasites of poultry. Of the fourteen families comprising the order Enoplida, only one family, Trichuridae, contains species parasitic in poultry.

The following key will aid in differentiating the eight families, containing species of poultry-parasitic nematodes.

1. Worms with free-living adult generation, that is, males and females developing outside of body; in digestive tract, hermaphroditic females only . . . . . . . . . . . . Strongyloididae
   Worms without a free-living generation, that is, incapable of producing males and females outside of body . . . . . . . . . 2
2. Worms hairlike or threadlike; esophagus tubular and capillary, the tube embedded in or otherwise in relation to a single row of cells; in crop and small intestine . . . . . . . . . . . . Trichuridae
   Worms thick as compared with above; esophagus well developed and muscular and with definite triangular lumen, not in relation to a single row of cells . . . . . . . . . . . . . . . 3
3. Cordons or other cephalic ornamentations present . . Acuariidae
   Cordons or other cephalic ornamentations absent . . . . . 4
4. Preanal sucker present . . . . . . . . . . Heterakidae
   Preanal sucker absent . . . . . . . . . . . . 5
5. Bursa present . . . . . . . . . . . . . . 6
   Bursa absent . . . . . . . . . . . . . . 7
6. Buccal capsule well developed and containing at least six teeth at base; oral opening hexangular . . . . . . . . Syngamidae
   Buccal capsule reduced and containing not more than three teeth at base or none . . . . . . . . . . . Trichostrongylidae
7. Pseudolabia absent . . . . . . . . . . Thelaziidae
   Pseudolabia present . . . . . . . . . . Spiruridae

**General morphology.** In general the body of a nematode is spindle-shaped with the anterior and posterior ends attenuated. The body covering or cuticle is usually marked by transverse grooves, and sometimes longitudinal folds or alae may be present. These alae may be confined to the anterior end of the body, in which case they are termed cervical alae; or they may be confined to the posterior part of the body, being then termed caudal alae. The latter are found on the tail of the male worm and, in the case of certain groups, are modified to form a bursa. Cuticular ornamentations are occasionally found on the anterior extremities of certain small groups of roundworms. These ornamentations may take the form of spines, cordons, or shields.

The mouth opening is located at the extreme tip of the anterior end of the body and is usually surrounded by lips bearing sensory organs. In the more generalized type of nematodes, the mouth leads directly into a mouth cavity. This cavity may be considerably reduced or absent in the more specialized groups of nematodes. Directly posterior to the mouth cavity is the esophagus. This part of the intestinal tract may be simple, i.e., consisting of one undivided part, or it may be more complex, consisting of a short, anterior muscular part and a long posterior glandular part. A bulb

may or may not be present at the posterior end of the esophagus. Following the esophagus is the intestine which is connected with the anal or cloacal opening in the posterior end of the body by a short rectum.

The nematodes are, with very few exceptions, sexually distinct. The male can usually be distinguished from the female by the presence of two—sometimes only one—chitinous structures known as spicules which are located in the posterior end of the body. The spicules have been considered as intromittent organs for use during copulation. That the spicules do take an active part in copulation has been observed many times. They have been observed to withdraw and insert alternately over extended periods during coitus. It has been reported that the primary function of the spicules is to keep the vulva and vagina open and, to some extent, guide the sperm into the female. The female reproductive products are discharged through the vulva, the position of which varies considerably in different groups of nematodes.

Sexual dimorphism is remarkably demonstrated by some species of nematodes. One of the most striking examples of this interesting phenomenon is *Tetrameres americana,* a nematode occurring in the proventriculus of certain kinds of poultry. The globular-shaped females enter the glands of the proventriculus or glandular stomach when very young, and as they begin to swell with eggs, their bodies assume the shape of the lumen of the glands. The adult male worm of this species is very much smaller than the female and retains the usual elongated nematode shape and lives free in the lumen of the proventriculus.

Nematodes are found in a variety of locations within the bodies of their hosts. The eyes, air sacs, thoracic and abdominal cavities, and the tracheae are some of the unusual places that nematodes occur as parasites of avian hosts. The intestinal tract is, of course, the habitat of the largest number of species of roundworms.

**Development.** Nematodes have both a direct (monoxenous) and an indirect (heteroxenous) type of development. Those worms with life histories of the first type require no invertebrate intermediate hosts to complete their life cycles and constitute approximately one-third of all the nematode species infesting poultry. However, the majority of the species of roundworms found in poultry are of the second type and depend upon such intermediate hosts as insects, snails, and slugs for the early stages of their development.

Regardless of the type of development that a certain species of nematode may have, it normally passes through four developmental stages before it becomes an adult; the latter is the final or fifth stage. Beginning with the second stage, each succeeding developmental stage in the life cycle is preceded by a molt. A molt is usually referred to as a shedding of the skin. In the case of some nematodes the loosened skin or cuticle may sometimes be retained for a short time as a protective covering, while in others it is shed at once.

Aside from the fact that certain nematodes require intermediate hosts to complete their development and others do not, the life histories of most nematodes infesting poultry are essentially the same. The eggs, which are deposited in the location in which the female worms are found, ultimately reach the outside in the droppings. This ex-corporal existence is apparently essential in order that the eggs may be rendered infective for the next host, be it avian or arthropod. The conditions existing within the body of the definitive host are usually inimical to the development of the eggs. However, when once outside the body of the bird host and in the presence of optimum moisture and temperature requirements, these eggs undergo development. The time required for the eggs to embryonate depends somewhat upon the species of parasite since, under similar environmental conditions, the eggs of some nematodes require only a few days to complete embryonation, while others require several weeks. In case of nematodes with a direct life cycle, the final host becomes infected by eating the embryonated eggs or the freed larvae. On the other hand, in the case of nematodes with an indirect life cycle, the intermediate host ingests the embryonated eggs or free larvae and retains the larvae within its body tissues. When a suitable final host eats the infested intermediate host containing infective larvae, it will become infected.

**Importance of nematodes of poultry.** Nematodes, as a whole, constitute the most important group of helminth parasites of poultry. Both in number of species affecting poultry and in the amount of damage done, this group of parasites far exceeds the flukes and cestodes.

Some of our most important individual worm parasites of poultry are found in this group. One species, *Heterakis gallinae*, plays an important role in the transmission of the protozoan disease known as blackhead. Although considered of little economic importance as parasites of the domestic fowl, this species apparently has caused serious and enormous losses to the poultry industry in the role of a carrier of the blackhead organisms. The gapeworm is perhaps the most serious nematode parasite of young poultry, particularly chickens and turkeys. Until recently, this worm was responsible for considerable losses among young birds both in this country and in Europe. Before changed poultry husbandry practices and other effective control measures reduced its devastating losses, this parasite was dreaded as much as blackhead. Despite all of our efforts to control this worm, serious outbreaks of epidemics among goslings and pheasants have recently been reported as due to the poultry gapeworm. Suffice to say, most of the nematodes parasitic in poultry and closely related birds may inflict serious injury to their respective hosts if infestations are sufficiently large.

The following is a list of species of nematodes found in poultry of this country, with their intermediate hosts, usual locations, and kinds of poultry affected.

| Nematodes | Location | Intermediate hosts | Definitive hosts |
|---|---|---|---|
| Oxyspirura mansoni | Eye | Cockroaches | Chicken, Turkey, Peafowl |
| Syngamus trachea | Trachea | None | Chicken, Turkey, Guinea fowl |
| Capillaria annulata | Esophagus, Crop | Earthworms | Chicken, Turkey, Guinea fowl |
| Capillaria contorta | Esophagus, Crop | None | Turkey, Duck, Bobwhite quail, Hungarian partridge, Ring-neck pheasant |
| Gongylonema ingluvicola | Crop | Unknown | Chicken, Turkey, Bobwhite quail |
| Dispharynx spiralis | Proventriculus | Sowbugs Pillbugs | Guinea fowl, Turkey, Chicken, Pigeon, Bobwhite quail |
| Seurocyrnea colini | Proventriculus | Cockroach | Turkey, Bobwhite quail, Sharp-tailed grouse, Prairie chicken |
| Tetrameres americana | Proventriculus | Grasshoppers Cockroaches | Chicken |
| Cheilospirura hamulosa | Gizzard | Grasshoppers Beetles Sandhoppers Weevils | Chicken, Turkey |
| Amidostomum anseris | Gizzard | None | Duck, Goose |
| Ascaridia galli | Small intestine | None | Chicken, Turkey |
| Ascaridia columbae | Small intestine | None | Pigeon |
| Ascaridia numidae | Small intestine | Unknown | Guinea fowl |
| Capillaria columbae | Small intestine | None | Pigeon, Chicken, Turkey |
| Capillaria longicollis | Small intestine | Earthworm | Chicken, Turkey |
| Ornithostrongylus quadriradiatus | Small intestine | None | Pigeon |
| Heterakis gallinae | Cecum | None | Chicken, Turkey |
| Strongyloides avium | Cecum | None | Chicken, Turkey |
| Trichostrongylus tenuis | Cecum | None | Chicken, Duck, Goose, Guinea fowl |
| Subulura brumpti | Cecum | Grasshoppers Beetles Mealworms | Chicken, Turkey, Bobwhite quail |
| Subulura strongylina | Cecum | Unknown | Chicken, Guinea fowl, Bobwhite quail |

## NEMATODES OF THE EYE

### THELAZIIDAE

The eyes of poultry are the seat of infection for a single species of nematode, *Oxyspirura mansoni*. This nematode belongs to the Thelaziidae. Members of this family have a buccal capsule which may be well developed or rudimentary, and the vulva is usually near the middle or posterior to middle of body.

### *Oxyspirura mansoni* (Cobbold, 1879)

**Synonym.** *Filaria mansoni* Cobbold, 1879.

**Description.** Mouth circular, surrounded by a 6-lobed chitinous ring
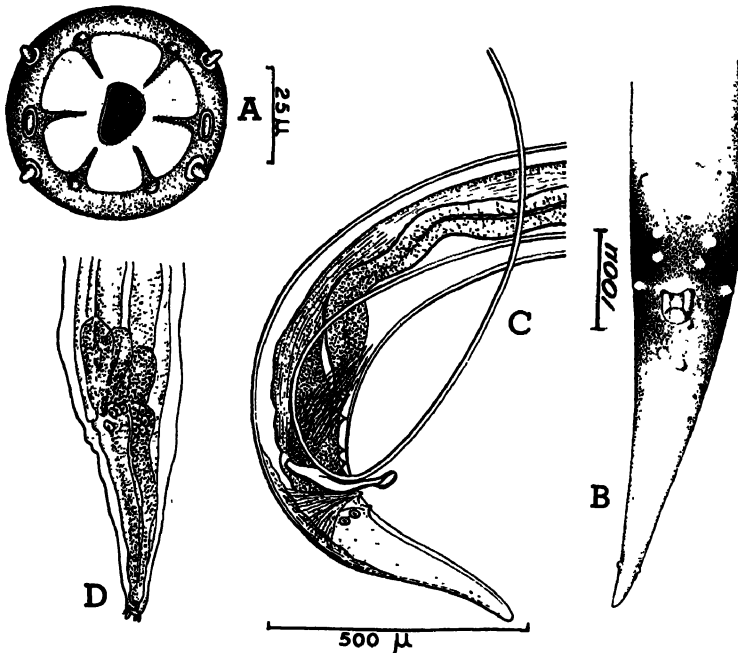


FIG. 32.1. *Oxyspirura mansoni.* A, front view of head; B, ventral view, and C, lateral view, of male tail. (After Ransom, 1904.) D, tail of second stage larva. (After Fielding, 1928.)

(Fig. 32.1A). Two pairs of subdorsal and 1 pair of subventral teeth in mouth cavity.

*Male* 8.2 mm. to 1.6 cm. long by 350μ wide. Tail curved ventrally, without alae. Four pairs of preanal and 2 pairs of postanal papillae (Fig. 32.1B). Spicules unequal (Fig. 32.1C); one is 3 to 4.55 mm. long and the other 180 to 240μ long.

*Female* 1.2 to 2 cm. long by 270 to 430μ wide. Vulva 780μ to 1.55 mm. from tip of tail. Eggs embryonated when deposited.

This roundworm is found beneath the nictitating membrane of poultry in the states of Florida and Louisiana.

**Life history.** Sanders (1929) found that an intermediate host was necessary for the successful transmission of this parasite from one bird host to another.

The complete life cycle of the eyeworm as worked out by Sanders is as follows: The eggs of the mature female worm are deposited in the eyes of the bird host. They are then washed down the tear ducts, swallowed, and passed to the exterior in the droppings. The cockroach, *Pycnoscelus* (*Leucophaea*) *surinamensis*, which is an omnivorous feeder, ingests the nematode eggs deposited in the droppings of infected birds. Within approximately 50 days following the ingestion of the infective eggs under experimental conditions, the cockroach contains in its body cavity mature larvae, which are capable of infecting a susceptible host. The mature larvae are often contained within cysts which are located deeply in the adipose tissue or along the course of the alimentary tract of the insect host. Some of the larvae release themselves from the capsules and are found free in the body cavity and legs of the cockroach. When an infected cockroach is swallowed by a chicken or other susceptible host, the infective larva is freed in the crop of the bird host, from which it later passes up the esophagus to the mouth and through the naso-lachrymal duct to the eye.

Experimental evidence indicates that various wild birds are capable of becoming infected with the eyeworm of poultry and, as a result, may serve as sources of infection for domestic birds. Such birds as the blackbird, *Agelaius phoeniceus*, the bobolink, *Dolichonyx oryzivorus*, the wild pigeon, *Columba livia*, the loggerhead shrike, *Lanius ludovicianus*, and the blue jay, *Aphelocoma cyanea*, have been experimentally infested with the eyeworm of poultry.

**Pathology.** Birds harboring eyeworms show a peculiar ophthalmia. They appear uneasy and continuously scratch at the eyes, which are usually watery and show much inflammation. The nictitating membrane becomes swollen and projects slightly beyond the eyelids at the corners of the eyes and is usually kept in continual motion as if trying to remove some foreign object from the eye. The eyelids sometimes become stuck together, and a white cheesy material collects beneath them. If left untreated, severe ophthalmia may develop, and as a result the eyeball may be destroyed. The worms are seldom, if ever, found in the eyes when severe symptoms are manifested, presumably due to unfavorable conditions existing there.

## NEMATODES OF THE RESPIRATORY TRACT
### SYNGAMIDAE

Roundworms, commonly designated as gapeworms, inhabit the trachea of young chickens, turkeys, and guinea fowls (Fig. 32.2). They are the cause

of the disease known as "gapes." The gapeworm belongs to the family Syngamidae, subfamily Syngaminae. Members of this subfamily are characterized by having the vulva in the anterior part of the body, with stoma well developed and hexagonal in cross section, and with corona radiata reduced or absent.

<center>*Syngamus trachea* (Montagu, 1811)</center>

**Synonyms.** *Fasciola trachea* Montagu, 1811; *Syngamus trachealis* Siebold, 1836.

**Description.** Red worms, the color more pronounced in female. Mouth



FIG. 32.2. *Syngamus trachea.* Trachea showing attached gapeworms.

orbicular, with a hemispherical chitinous capsule, usually with 8 sharp teeth at the base. Mouth surrounded by a chitinous plate, the outer margin of which is incised to form 6 festoons opposite each other. Male permanently attached in copula to female, forming a Y (Fig. 32.3B).

*Male* 2 to 6 mm. long by 200$\mu$ wide. Bursa obliquely truncated, provided with rays, sometimes with strikingly asymmetrical dorsal rays. Spicules equal, slender, short, 57 to 64$\mu$ long.

*Female* 5 mm. to 2 cm. long (longer in the turkey) by 350$\mu$ wide. Tail end conical, bearing a pointed process. Vulva prominent, about one-fourth of body length from anterior end, but the position varies with age. Eggs 90$\mu$ long by 49$\mu$ wide, ellipsoidal, operculated (Fig. 32.3A).

The gapeworm, *Syngamus trachea*, is sometimes designated as the "red-worm" or "forked-worm" because of its red color and because the male and female are joined together so that they appear like the letter Y. This parasite is cosmopolitan in distribution.

**Life history.** The life history of the gapeworm is peculiar in that transmission of this parasite from bird host to bird host may be successfully accomplished either directly by the feeding of embryonated eggs or infective larvae, or indirectly by the ingestion of earthworms containing free or encysted gapeworm larvae which they had obtained by feeding on contaminated soil. The female gapeworm deposits its eggs through the vulvar
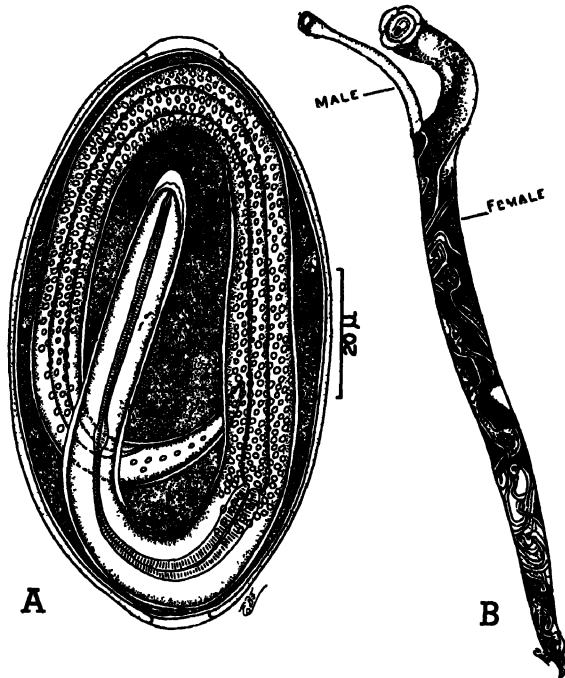


FIG. 32.3. *Syngamus trachea.* A—enlarged egg showing fully developed embryo. B—drawing of male and female gapeworms.

opening underneath the bursa of the attached male into the lumen of the trachea. The eggs reach the mouth cavity, are swallowed, and pass to the outside in the droppings. Following a period of incubation of approximately 8 to 14 days under optimum conditions of moisture and temperature, these eggs become embryonated. Soon after embryonation, some of the eggs may hatch, the larvae living free in the soil. Should specimens of the earthworms, *Eisenia* (H) *foetidus* and *Allolobophora* (H) *caliginosus*, and perhaps others, be present in the soil which has been contaminated with feces containing the eggs of gapeworms, these annelids will become infected with

gapeworm larvae. Within the earthworm, the larvae penetrate the intestinal wall, enter the body cavity, and finally invade the body musculature in which they may encyst for an indefinite period. Taylor (1938) stated that gapeworm larvae may remain infective to young chickens in the earthworm for as long as four and one-third years. This author also found that slugs and snails may also serve as transfer hosts of *Syngamus trachea* larvae and that live gapeworm larvae were obtained from snails over a year after infection. These mollusks are not essentially true intermediate hosts in the strict sense of the word, since they are not absolutely necessary for the successful transfer of the gapeworm from one susceptible bird host to another.

Clapham (1934) and other investigators have observed that strains of *Syngamus trachea* taken from various wild and domestic birds were more readily transferred to young chickens and with a greater degree of success if the earthworm was employed as an intermediary.

The exact path of migration of the gapeworm larva after it has once entered the intestinal tract of the avian host until it reaches the lung is not known at the present time. Walker (1886) believed that the larvae, after being swallowed by the definitive host, passed through the esophageal wall and entered the lungs directly from the outside. More recent observations have indicated that the path of migration may be via the blood stream. However, convincing evidence which tends to show the true path of migration is still lacking.

Observations by Wehr (1937b) have shown that the infective larvae reach the lungs in an apparently unchanged condition within at least 6 hours after they have been ingested by the bird. By the third day following inoculation, the larvae have developed to the fourth stage, and by the seventh day several fourth-stage larvae and a few immature adults—one pair of the latter in copula—were found in the lungs; five pairs of immature adults in copula were found in the trachea. It is evident from these and other observations that the male and female of *Syngamus trachea* copulate as young adults while in the lungs sometime between the third and seventh days following infection, and that the worms reach the trachea about the seventh day after ingestion of the embryonated eggs and larvae. These findings differ from those of Ortlepp (1923) who believed that the fourth stage was the final stage in the development of this nematode, and that the second-stage larvae represented the infective stage. This latter observation is obviously an error, since the gapeworm embryo has been observed to molt twice inside the egg.

Approximately two weeks are required for the infective larvae to reach sexual maturity and for eggs to appear in the droppings. About one-half of this time is spent in the lungs and the other half in the trachea.

The role played by the wild birds in the spread of gapeworm disease is

still undecided. So far as it is known at the present time, wild birds probably are not an important factor in the spread of gapeworm disease in this country.

**Pathology.** Young birds are most seriously affected with gapeworms. The rapidly growing worms soon obstruct the lumen of the trachea and cause the birds to suffocate and die. Turkey poults, baby chicks, and pheasant chicks are very susceptible to infection with gapeworms, whereas the young of the other species of poultry which have been experimentally inoculated with the infective eggs and larvae of gapeworms are not so seriously affected. Turkey poults usually develop gapeworm symptoms earlier and begin to die sooner following gapeworm infection than young chickens. Experimentally infested guinea fowls, pigeons, and ducks do not exhibit characteristic symptoms of gapeworm infestations. Young pheasants, however, suffer from the disease to an extent comparable to that of young chicks and turkey poults. Full-grown birds rarely show characteristic gapeworm symptoms, unless heavily infested. Investigators in this country have indicated that chickens ten weeks or older are very difficult to infect experimentally with gapeworms. However, Crawford (1940) reported that gapeworms occurred commonly in the tracheae of fowls of all ages, even in three-year-old hens, in Ceylon. He stated that the number of worms found in the trachea of each fowl was usually small and that adult hens not infrequently were seen to exhibit typical symptoms of gapeworm diseases. He considered the adult fowl to be an important factor in the perpetuation of gapeworm disease in Ceylon. Olivier (1943) reported the occurrence of *Syngamus trachea* in mature chickens in Maryland. One of these birds was heavily infected and exhibited typical clinical symptoms.

Young birds infested with gapeworms show symptoms of weakness and emaciation, and usually spend much of their time with the eyes closed and the head drawn back against the body. From time to time they throw their heads forward and upward, and open the mouth wide in order to draw in air. It is not an uncommon occurrence to see an infested bird give its head a convulsive shake in an attempt to remove the obstruction from the trachea so that normal breathing may be resumed. Little or no food is taken by birds in the advanced stages of infestation, and death is usually the end result.

An examination of the tracheae of infested birds shows that the mucous membrane is extensively irritated and inflamed; coughing is apparently the result of this irritation to the mucous lining. Lesions are usually found to be present in the tracheae of turkeys and pheasants, but seldom if ever seen in the tracheae of young chickens and guinea fowls. Observations have shown that these lesions or nodules are produced as a result of an inflammatory reaction set up at the site of the attachment of the male worm. Since lesions

have been observed only at the point of attachment of the male worm and observations have shown that the head of the male is deeply embedded in the nodular tissue, it is, therefore, believed that the male worm usually remains permanently attached to the tracheal wall throughout the duration of its life. The female worms apparently detach and reattach from time to time in order to obtain a more abundant supply of food.

It must be remembered, however, that there are other diseases which may cause birds to gape, such as bronchitis and laryngotracheitis. In order that one may be sure just what is the cause of the gaping, it is necessary to make a post-mortem examination of one or more of the affected birds. If gapeworms are not present in the trachea, bronchitis, laryngotracheitis or some other disease causing symptoms similar to gapeworm disease must be diagnosed.

## NEMATODES OF THE ALIMENTARY TRACT
### CROP

At least three species of nematodes, commonly referred to as crop worms, occur in the crop of domestic fowls. Two of these are commonly known as capillarid worms or hairworms and belong to the family Trichuridae, while the third is designated as the gullet worm and is a member of the family Thelaziidae.

### TRICHURIDAE

The members of this family are characterized by having the body more or less clearly divided into an esophageal portion and a posterior portion which contains the other organs. The esophagus is a cuticular tube embedded in one side of a single or double row of esophageal glands. The male possesses only a single spicule. Vulva is located at junction of esophageal and posterior portions of body.

### *Capillaria annulata* (Molin, 1858)

**Synonyms.** *Trichosoma annulatum* Molin, 1858; *Thominx annulata* (Molin, 1858) Cram, 1925.

**Description.** Worms long and threadlike. Cuticle much enlarged just behind head in adult specimens to form a bulbous swelling (Fig. 32.4A). Not far behind this, the cuticle is thrown into wavy transverse folds for a short distance.

*Male* usually 1 to 2.5 cm. long by 52 to 74μ wide. Tail ends in two inconspicuous round lateral flaps, united dorsally by a cuticular flap. Spicule sheath beset with fine spines (Fig. 32.4B). Spicule is said to be lacking.

*Female* usually 2.5 to 6 cm. long by 77 to 120μ wide. Posterior portion of body (posterior to vulva) about 7 times as long as anterior portion. Vulva circular, located about opposite the termination of the esophagus. Eggs operculated, 55 to 66μ long by 26 to 28μ wide.

*Capillaria annulata* occurs naturally in the bobwhite quail, domestic chicken and turkey, pheasant, and Hungarian partridge. Worms of this species are similar in appearance to those of *Capillaria contorta* except that they are shorter and possess a cuticular swelling just back of the head.

**Life history.** Eggs of this parasite pass out in the droppings of the infested birds. They develop very slowly; a period of 24 days to over one month is sometimes necessary before they have reached the stage at which they contain an active embryo. Wehr (1936) discovered that the earthworm is required in order to successfully transmit *C. annulata* from one bird host to another. He demonstrated that under both natural and artificial conditions two species of earthworms, *Eisenia* (H) *foetidus* and *Allolobophora* (H) *caliginosus*, served as intermediate hosts of this crop worm. Chickens and other susceptible hosts become infected with this crop worm by swallowing infested earthworms.

**Pathology.** Cram (1926c) reported this worm as being associated with the deaths of turkeys in Maryland. The habit of burrowing into the crop mucosa causes a thickening of the crop wall and an enlargement of the glands in the areas in which the worms are



Fig. 32.4. *Capillaria annulata*. A—head end. B—male tail. (After Ciurea, 1914.)

located. Usually there is a slight or severe inflammation of the crop and esophageal walls, depending upon the severity of the infestation. In heavy infestations, the inner surface of the crop is immensely thickened, roughened, and badly macerated, the masses of worms concentrated primarily in this sloughing tissue (Fig. 32.5).

In pheasants, quail, and other gallinaceous game birds, infestations with this parasite often prove fatal. In these birds the symptoms reported are principally malnutrition and emaciation, associated with severe anemia. Allen and Gross (1926) reported severe anemia in an infested ruffed grouse, shortly before death.

Hung (1926) made a histopathological study of three cases of varying intensity and reported the following changes: "On the basis of the above

observations it is quite evident that the pathological changes caused by
*C. annulata* may be divided into three stages. The first stage is the hyperemic
stage in which only hyperemia and lymphocytic infiltrations are present.
In the second stage the yellowish-white nodules are present, and the lym-
phatic apparatus is enlarged and sometimes necrotic. The enlargement of



Fig. 32.5. *Capillaria annulata.* Damage done to crop of quail, as compared with the
thin-walled normal crop.

the lymph follicles gives the appearance of nodules. The third stage is that
of the formation of the pseudomembrane, in which the mucosa is covered
with a membrane containing fibrin."

*Capillaria contorta* (Creplin, 1839)

**Synonyms.** *Trichosoma contortum* Creplin, 1839; *Thominx contorta*
(Creplin, 1839) Travassos, 1915.

**Description.** Body threadlike, attenuated anteriorly and posteriorly.

*Male* 8 mm. to 1.7 cm. long by 60 to 70μ wide. Two terminal latero-
dorsal prominences on tail end. Spicule very slender and transparent, about
800μ long, according to Travassos. Spicule sheath covered with fine hair-
like processes (Fig. 32.6B).

*Female* 1.5 to 6 cm. long by 120 to 150μ wide. Vulva prominent, circular,
140 to 180μ posterior to beginning of intestine (Fig. 32.6A).

*Capillaria contorta* has been reported from a large number of hosts, in-

cluding the duck, turkey (both domestic and wild), pheasant, quail, and ruffed grouse.

**Life history.** Eggs are apparently deposited in tunnels in the crop mucosa and escape into the lumen of crop and esophagus with the sloughed mucosa. They are found abundantly in droppings from infested birds. Approxi-
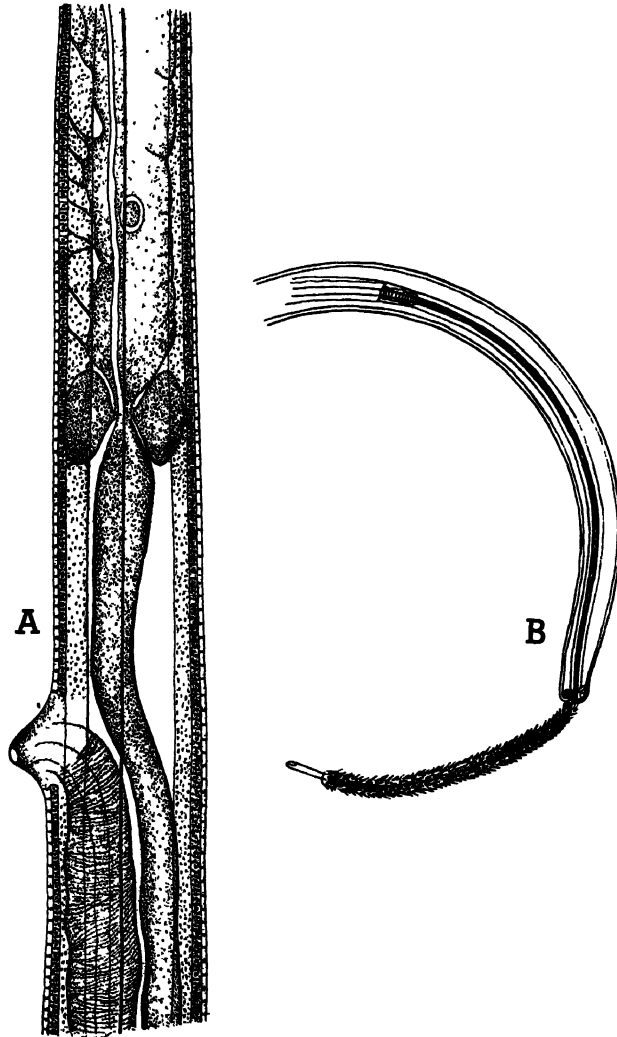


FIG. 32.6. *Capillaria contorta.* A—region of vulva. (After Eberth, 1863.) B—male tail. (After Travassos, 1915.)

mately one month or slightly longer is required for embryos to develop within the eggs. Worms mature and eggs pass to the outside in the droppings

of susceptible avian hosts in from one to two months after feeding the
embryonated eggs. Attempts to experimentally infect chickens, guinea
fowls, and pigeons were unsuccessful.

**Pathology.** When present in large numbers these worms are highly dan-
gerous. In light infestations the wall of the crop and esophagus become
slightly thickened and inflamed. In heavy infestations, there is a marked
thickening and inflammation, with a flocculent exudate covering the mucosa
and with more or less sloughing of the mucosa (Fig. 32.7).



FIG. 32.7. Section of crop of bobwhite quail, showing *Capillaria contorta* and damage
produced by it. Experimental infestation. ×114.

Affected birds become droopy, weak, and emaciated. Many deaths due
to infestation with this worm have been observed among wild turkeys and
Hungarian partridges in this country.

The author recently visited a flock of turkeys in Virginia, among which
several birds were reported to have died from heavy infestations with this
crop worm. A number of visibly affected birds had been segregated from the
main flock and were held in a pen by themselves. These birds moved only
when disturbed, and then very slowly and with an unsteady gait. Occa-
sionally a bird was seen to fall back on its hock joints and assume a penguin-
like position. Others extended and retracted their heads and necks as if
attempting to relieve an obstruction in their throats. The crops of the most
severely affected birds were filled with a fetid liquid. Emmel (1939) ob-
served that infestation with this crop worm appeared first in the older birds,
and later those of all ages became affected. Affected birds appeared indis-
posed, weak, and droopy, with the forepart of the body slightly elevated.
The birds were not inclined to move unless forced to do so. He also observed
that the birds occasionally assumed a penguin-like posture, with the head

drawn close to the body, and that affected birds frequently swallowed and in doing so always extended and "ducked" their heads.

THELAZIIDAE

For family diagnosis, see page 764.

*Gongylonema ingluvicola* (Ransom, 1904)

**Description.** Anterior end of body with a zone of shieldlike markings, few and scattered near head, numerous, and arranged in longitudinal rows farther back (Fig. 32.8A).

*Male* 1.7 to 2 cm. long by 224 to 250μ wide. Cervical papillae about 100μ from head end. Tail with two narrow bursal asymmetrical membranes. Genital papillae variable in number and asymmetrical; preanal papillae up to 7 on left side and up to 5 on the right side (Fig. 32.8 B). Left spicule as long, or nearly as long (1.7 to 1.9 cm.), as body and 7 to 9μ wide, with a barbed point; right spicule 100 to 120μ long and 15 to 20μ wide.

*Female* 3.2 to 5.5 cm. long by 320 to 490μ wide. Vulva 2.5 to 3.5 mm. from tip of tail.

This worm has been reported from the chicken, turkey, and quail in the United States.

**Life history.** Cram (1931a) fed larval roundworms collected from the beetle, *Copris minutus*, to a chicken and recovered a single male



FIG. 32.8. *Gongylonema ingluvicola.* A— head. B—male tail. (After Ransom, 1904.)

specimen of a species of *Gongylonema*, tentatively identified as *G. ingluvicola*. Cram subsequently infected cockroaches by feeding embryonated eggs of *G. ingluvicola* derived from mountain quail. Some of the larvae recovered from the cockroaches were fed to a chicken. No worms were found when the chicken was killed 79 days later.

**Pathology.** The only damage that has been associated with these worms is the local lesions in the form of burrows in the mucosa of the crop.

GENERAL DISCUSSION

Crop worms penetrate into the mucosa of the crop and esophagus and make tortuous burrows in which they live. Wehr (1937a) observed that

each of these species of crop worms, when seen in their normal positions in the mucosa, displayed a different body contour. The value of this observation lies in the fact that identifications of the worms involved may be made with a reasonable degree of accuracy without resorting to the time-consuming task of making a detailed microscopic examination of each worm. In situ, all three species of worms assume a folded position, differing, however, in the character of the folds. In case of the so-called gullet worm, *Gongylonema ingluvicola*, the perspective is one of a series of folds approximately uniform in size and shape, following one another in close succession and usually extending in a straight line. The body shape of the two species of *Capillaria* consist of a series of irregularly shaped folds. In these cases, however, the size of the worms becomes the deciding factor. *Capillaria annulata* may be readily differentiated from *Capillaria contorta* by its much smaller size. In case of any doubt as to the identity of the two species, the specimens may be removed from their burrows and examined under the microscope. Should a cuticular swelling be present directly back of the head the worm is *C. annulata;* if this structure is absent, it is *C. contorta*.

*Capillaria contorta* is frequently found in large numbers in wild and domestic birds. In the United States, this crop worm has been reported to occur naturally in the Hungarian partridge, wild turkey, ring-necked pheasant, California valley quail, wild and domestic ducks, the domestic turkey, and possibly others. This species is the longest of the three species of crop worms.

## STOMACH

Nematodes inhabiting the proventriculus of domestic poultry belong to two families, namely, Acuariidae and Spiruridae.

### ACUARIIDAE

The acuariids are characterized by having well-developed pseudolabia and cuticular ornamentations on the anterior part of the body.

### *Dispharynx nasuta* (Rudolphi, 1819)

**Synonyms.** *Spiroptera nasuta* Rudolphi, 1819; *Dispharagus spiralis* Molin, 1858; *Acuaria spiralis* (Molin, 1858) Railliet, Henry, and Sisoff, 1912. Goble and Kutz (1945) concluded that all the forms of Dispharynx which have been recently recorded from galliform, columbiform, and passiform birds in the Western Hemisphere are conspecific and that a morphological study of these forms leads them to consider their identity as *Dispharynx nasuta* (Rudolphi, 1819) Stiles and Hassall, 1920. *Dispharynx nasuta* (Rudolphi, 1819) Stiles and Hassall, 1920, has priority over *Dispharynx spiralis* (Molin, 1858) Skrjabin, 1916.

**Description.** Four wavy cuticular cordons on anterior end, originating at base of lips, recurrent, the distal extremity of the cordons turning forward

and extending anteriorly a short distance (Fig. 32.9A). Postcervical papillae small, bicuspid, situated between the recurrent branches of the cordons. Body usually rolled in a spiral (Fig. 32.9B).

*Male* 7 to 8.3 mm. long by 230 to 315µ wide. Five pairs of postanal and 4 pairs of preanal papillae (Fig. 32.9C). Long spicule 400µ long, slender and curved; short spicule 150µ long, navicular.
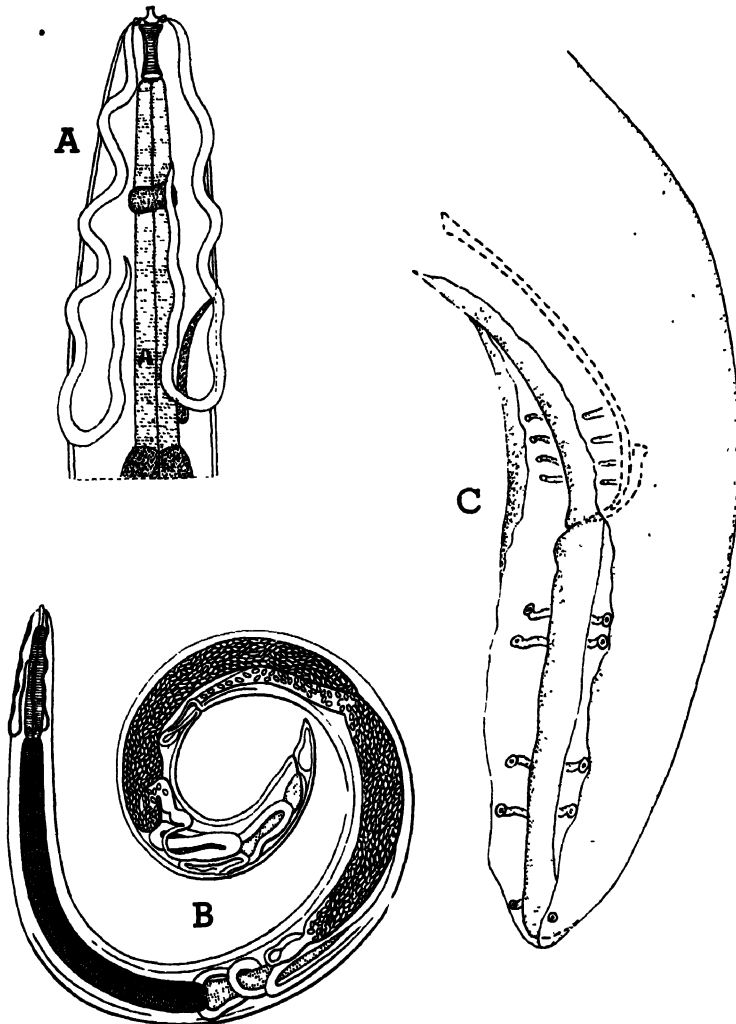


Fig. 32.9. *Dispharynx nasuta.* A—head end. (After Seurat, 1915.) B—female, enlarged. (After Piana, 1897.) C—male tail. (After Cram, 1928.)

*Female* 9 to 10.2 mm. long by 360 to 565µ wide. Small mucron on tip of tail. Vulva in posterior portion of body. Eggs embryonated when oviposited.

This parasite has been encounterd in the proventriculus of the chicken,

turkey, guinea fowl, pigeon, pheasant, ruffed grouse, bobwhite quail, Hungarian partridge, and other gallinaceous birds. In addition, it has been found in a number of passerine birds in the United States. Yeatter (1934) found the incidence of this parasite among Hungarian partridge in the Great Lakes region to be 31.6 per cent. Bump (1935) stated that this worm was the most important parasite recovered from the ruffed grouse in New York State.

**Life history.** The pillbug, *Armadillidium vulgare*, and the sowbug, *Porcellio scaber*, were demonstrated to serve as intermediate hosts in experimental infections by Cram (1931b). The writer has repeatedly confirmed Cram's studies, using pigeons as definitive hosts. Within 4 days after the ingestion of the embryonated eggs by these isopods, the larvae have escaped from the eggs and are found among the tissues of the body cavity of the crustacean. The larva completes its development in the isopod within approximately 26 days; it has then reached the third or infective stage.

The definitive host becomes infected with the above nematode by swallowing infested pillbugs or sowbugs with the food or water. According to Cram (1931b) the female worms become sexually mature and are depositing eggs 27 days after ingestion by a susceptible vertebrate host.

**Pathology.** These roundworms are usually seen with their heads buried deeply into the mucosa. The formations of ulcers are often observed in the proventriculi of infested birds. In case of heavy infestations, the wall of the proventriculus becomes tremendously thickened and macerated, tissue layers are indistinguishable, and the parasites become almost completely concealed beneath the proliferating tissue.

Allen (1925) believed that *D. nasuta* (*D. spiralis*) was the chief cause of "Grouse Disease" in northeastern United States. Heavy infestations of this parasite resulted in the death of many carrier pigeons of the Signal Corps of the United States Army, Fort Sam Houston, Texas, a number of years ago, according to Cram (1928). Several wild pigeons trapped at the Balboa Zoological Park, San Diego, California, and examined by the writer were found to be heavily infested with this parasite.

### *Seurocyrnea colini*

*Seurocyrnea colini* is of common occurrence in the bobwhite quail of the southeastern states and has occasionally been collected from this same host and closely related birds in some of the northeastern states. It has also been reported from the turkey in Georgia, the prairie chicken in Wisconsin, and the sharp-tailed grouse in Wisconsin and Montana.

The preferred location of this nematode is in the wall of the proventriculus at its junction with the gizzard. The slender, yellowish-white worms are similar in appearance to *Cheilospirura hamulosa*, but are smaller and lack the so-called cordons or cuticular ornamentations on the anterior part of

the body. The head structures are quite complicated, and the tail of the male has winglike expansions or alae (Fig. 32.10 A, B, C, D, and E).

The life history of this nematode is indirect, requiring the cockroach, *Blatella germanica,* as a temporary host. Since this intermediate host has been incriminated in an experimental role only, it is not known whether it actually serves in this same capacity under natural conditions.
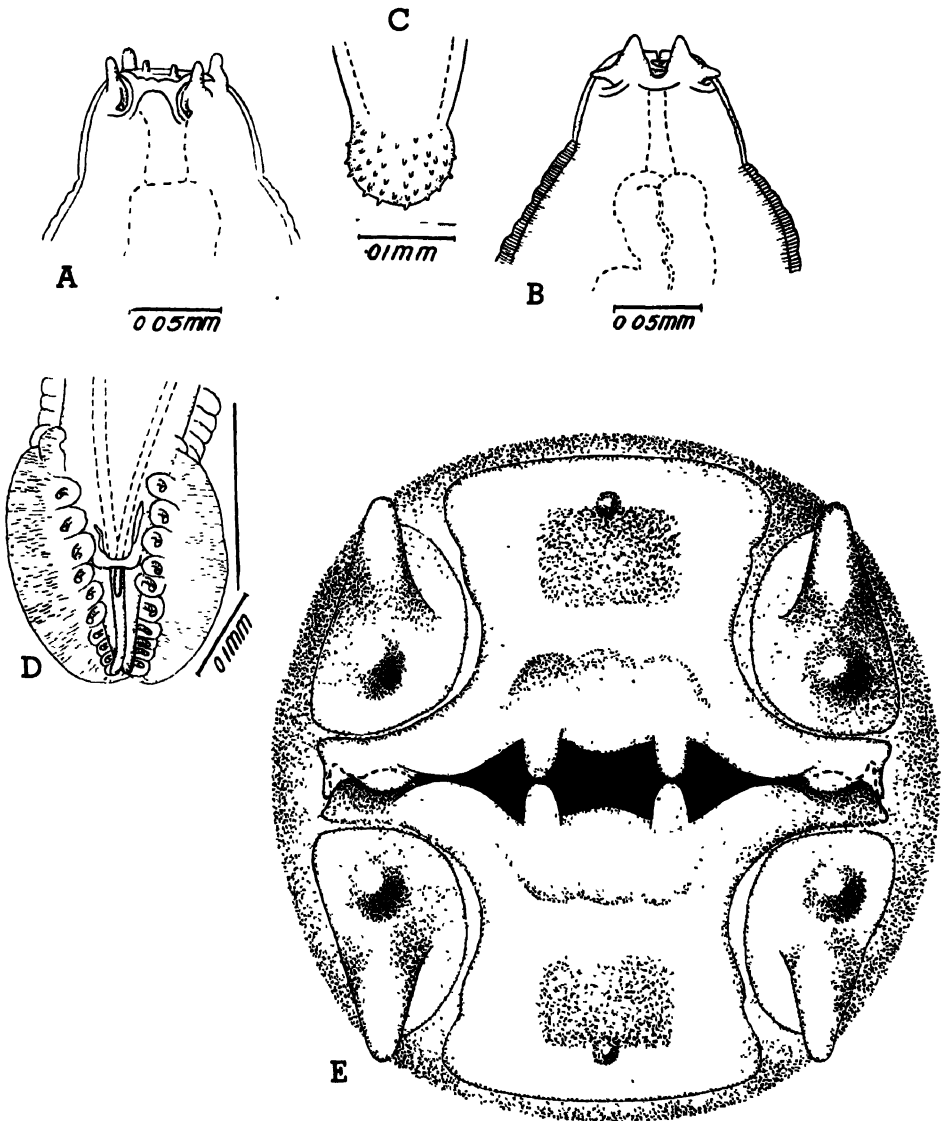


Fig. 32.10. *Seurocyrnea colmi.* A—head, oblique lateral view. B—head, ventral view. C—tail of third-stage larva. D—male tail. E—head, *en face* view, semidiagrammatic. (After Cram, 1927.)

There has been little or no pathological change observed in connection with infestations of this parasite.

The spirurids are characterized by having well-developed pseudolabia, cephalic papillae usually posterior to pseudolabia, and interlabia present or absent. The only member of this family found in poultry of this country have interlabia, and the sexes are distinctly dimorphic.

### *Tetrameres americana* Cram, 1927

**Description.** Mouth surrounded with 3 small lips; buccal cavity present.

*Male* 5 to 5.5 mm. long by 116 to 133µ wide. Two double rows of posteriorly directed spines extend throughout whole body length, in the submedian lines. Cervical papillae present. Tail long and slender. Two unequal spicules, 100µ and 290 to 312µ long, respectively.

*Female* 3.5 to 4.5 mm. long by 3 mm. wide. Body globular, blood red in color, with 4 longitudinal furrows (Fig. 32.11). Uteri and ovaries very long, their numerous coils filling the body cavity.

The female of this species occurs in the glandular stomachs of the chicken and bobwhite quail (Fig. 32.12). At necropsy, these bright red worms are often observed through the wall of the unopened proventriculus. The male of this species is very small, almost microscopic in size, and resembles other nematodes in shape. It has never been observed elsewhere than on the surface of the mucosa of the proventriculus. However, the males of some of the species of Tetrameres occurring in wild birds have been found on several occasions together with the females in the same glands. From all indications, it seemed that the two sexes, in the cases cited, were permanent residents of the glands in which they were found. If the male of *Tetrameres americana* enters the glands of the proventriculus, it apparently does so only long enough to mate with the female.



FIG. 32.11. *Tetrameres americana.* Enlarged drawing of female (original).

Cram (1931a) found this parasite to be common in quail which had been raised in captivity and in close proximity to poultry in Virginia. Stoddard (1931) reported *Tetrameres americana* as being found occasionally in quail

captured in its natural habitat in the southeastern part of the United States.

Swales (1933) described *Tetrameres crami* from the proventriculus of a domestic duck in Canada. He stated that this species, of which the female only is known, differs from *Tetrameres americana* chiefly in the shorter muscular esophagus and the relative positions of the anus and vulva.

Another species of Tetrameres, *Tetrameres fissispina,* a species closely



Fig. 32.12. *Tetrameres americana.* Proventriculus showing female worms in glands. (After Cram, 1930.)

related to *T. americana,* has been reported from wild and domestic ducks and chickens in Europe. Sugimoto and Nishiyama (1937) stated that this roundworm was fairly common in chickens in Formosa.

**Life history.** Cram (1931b) discovered that *T. americana* required an intermediate host for its complete development. She fed embryonated eggs of this worm to two species of grasshoppers, *Melanoplus femurrubrum* and *M. differentialis,* and a species of cockroach, *Blatella germanica,* and recovered infective larvae from the body cavities of these insects in about 42 days after the ingestion of the eggs. When the grasshopper or cockroach is swallowed by a suitable bird host and digested in its stomach, the larvae escape; they remain on the surface of the proventriculus and develop into adults within a few days. The complete life cycle of *Tetrameres fissispina* involves such intermediate hosts as the amphipod, *Gammarus pulex;* the

cladoceran, *Daphnia pulex;* and several species of grasshoppers, cockroaches, and earthworms.

**Pathology.** According to Sugimoto and Nishiyama (1937), infested chickens become emaciated and anemic as a result of heavy infestations. Cram (1931a) reported that *Tetrameres americana* has not been observed to produce any damage in quail. Barber (1916) stated that this proventricular worm was the cause of a serious catarrhal condition in chickens in Guam. In his report, he mentioned that the walls of the proventriculus were so thickened that the lumen was almost entirely obliterated; as many as forty-seven worms were found embedded in the wall.

## GIZZARD

Gizzard nematodes belong to two families, namely, Acuariidae and Trichostrongylidae.

### ACUARIIDAE

For family diagnosis, see page 776.

#### *Cheilospirura hamulosa* (Diesing, 1851)

**Synonyms.** *Spiroptera hamulosa* Diesing, 1851.

**Description.** Two large, triangular, lateral lips. The 4 cuticular cordons double, irregularly wavy, and extending almost to posterior extremity; not anastomosing or recurring anteriorly (Fig. 32.13A).

*Male* 9 mm. to 1.9 cm. long. Spicules very unequal and dissimilar, the left long and slender, the right short and curved. Tail tightly coiled; 2 very wide caudal alae present. Ten pairs of caudal papillae (Fig. 32.13B).

*Female* 1.6 to 2.5 cm. long. Vulva slightly posterior to middle of body. Tail pointed. Eggs embryonated when deposited.

This roundworm occurs commonly underneath the horny lining of the gizzard near the openings of the proventriculus and intestine in chickens and has occasionally been reported from the same locations in turkeys. It is widely distributed in the United States.

**Life history.** Investigations have shown that grasshoppers, beetles, weevils, and sandhoppers serve as intermediate hosts of *C. hamulosa* under natural as well as experimental conditions. Chickens and other susceptible avian hosts become infested with the adults of this roundworm by ingesting grasshoppers, beetles, weevils, and sandhoppers which are infested with larvae of this worm.

The infective or third stage larva may be recognized easily by the 2 prominent liplike structures at the anterior end of the body, the dorsal curvature of the posterior portion of the body, and the presence of four digitiform processes at the tip of the tail.

**Pathology.** When present in small numbers, these worms cause no evident effect on the health of the birds. In such infestations the lining of the gizzard may show small local lesions which may also involve the muscular tissue.

Soft nodules enclosing parasites may be found in the muscular portion of the gizzard. In heavy infestations, the wall of the gizzard may be seriously damaged. Le Roux (1926) reported that this parasite may weaken the wall to such an extent as to cause it to rupture, with ultimate formation of a sac or pouch.

### TRICHOSTRONGYLIDAE

Members of this family are characterized by having reduced or rudimentary mouth cavity, corona radiata absent, and usually a well-developed bursa.

*Amidostomum anseris* (Zeder, 1800)

**Synonyms.** *Strongylus anseris* Zeder, 1800 in part; *Amidostomum nodulosum* (Rudolphi, 1803) Seurat, 1918.

**Description.** Worms slender and reddish. The short wide buccal capsule has 3 pointed teeth at its base (Fig. 32.14A).

*Male* 10 to 17 mm. long by 250 to 350μ wide. Bursa with 2 large lateral lobes and a small median lobe (Fig. 32.14C). Dorsal ray short, bifurcating posteriorly and the bifurcations forked and terminating in two tips. Spicules 200μ long, slender, and cleft near their middle. Gubernaculum slender and 95μ long.

*Female* 12 to 24 mm. long, 300 to 400μ wide at vulva, thinning toward both extremities. Vulva transverse, in posterior part of the body (Fig. 32.14B). Eggs thin-shelled.

This worm occurs very commonly underneath the horny lining of the gizzard of wild ducks and geese. In the United States it has been reported from domestic geese in the states of New York and Washington. No doubt this parasite has a much wider distribution in this country than the present records indicate.



FIG. 32.13. *Cheilospirura hamulosa.* A—head. (After Drasche, 1884.) B—male tail. (After Cram, 1931.)

**Life history.** Eggs pass out in the droppings of infested birds in a partly developed stage, active embryos developing within a few hours and hatching taking place within a few days. Susceptible bird hosts become infected by swallowing with their food or drinking water these infective larvae. Adult worms are recovered within approximately 40 days after the feeding of infective larvae.

**Pathology.** In the United States, Cram (1926a) reported an outbreak of

amidostomiasis in a flock of geese in New York in which a large number of deaths occurred. Heavy losses among geese in Europe have been attributed to this nematode. Young birds show symptoms of loss of appetite, dullness, and emaciation. At necropsy, the lining of the gizzard of a heavily parasitized bird appears necrotic, loosened, often sloughed in places, and is dark brown or black in color in areas adjacent to the cite of the worms.

Bunyea and Creech (1926) found a very noticeable leukocytic invasion of the mucosa propria, with eosinophilic cells strikingly predominant.

## INTESTINAL TRACT

### HETERAKIDAE

The members of this family are characterized by having 3 prominent lips, valvulated bulb present or absent, and preanal sucker present or absent.
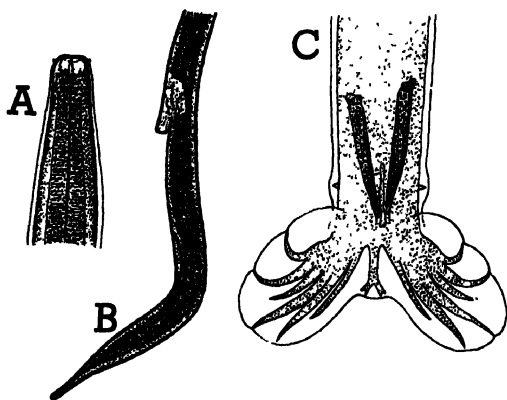


FIG. 32.14. *Amidostomum anseris.* A—head end. (After Railliet, 1893.) B—vulva and tail of female. (After Reinhardt, 1922.) C—male tail. (After Railliet, 1893.)

*Ascaridia galli* (Schrank, 1788)

**Synonyms.** *Ascaris galli* Schrank, 1788; *Heterakis lineata* Schneider, 1866; *Heterakis inflexa* (Zeder, 1800) Schneider, 1866.

**Description.** Worms large, thick, yellowish white (Fig. 32.15). Head with 3 large lips. Valvulated bulb and preanal sucker slightly salient.

*Male* 5 to 7.6 cm. long by 490μ to 1.21 mm. wide. Preanal sucker oval or circular, with strong chitinous wall with a papilliform interruption on its posterior rim. Tail with narrow caudal alae or membranes, and 10 pairs of papillae. Spicules equal and narrow.

*Female* 6 to 11.6 cm. long by 900μ to 1.8 mm. wide. Vulva in anterior part of body. Eggs elliptical, thick-shelled, not embryonated at time of deposition (Fig. 32.16).

This large roundworm is one of the most common nematode parasites of the chicken in the United States and elsewhere. It occurs occasionally in turkeys, but no serious pathologic effects have been reported from its presence in that host.

Specimens of this parasite have been recovered on a number of occasions from broken eggs. The worms had presumably wandered up the oviduct

from the intestine via the cloaca with subsequent inclusion in the developing egg.

**Life history.** The life history is simple and direct. According to Itagaki (1927), the infective eggs which are swallowed by the susceptible host hatch either in the proventriculus or in the duodenum. Ackert (1931) observed that the young larvae, after hatching from the eggs, live free in the lumen of the posterior portion of the duodenum for the first 9 days, following which they penetrate the mucosa and cause hemorrhages. The young worms have again entered the lumen of the duodenum by the seventeenth or eighteenth day and remain there until maturity, which is reached within approximately 50 days after the ingestion of the embryonated eggs.

Under optimum conditions of temperature and moisture, the eggs in the droppings will develop to infectivity in 10 to 12 days; under less favorable conditions a longer time is necessary. The eggs are quite resistant to low temperatures. Ackert found that in the early stages of development, eggs survived freezing at —12° to —8° C. for 15 hours, but not for 22 hours; fertile eggs kept at 0° C. for one month were unable to reach the infective stage subsequently, whereas eggs from the same culture kept concurrently at 10° C. for a month, developed normally to the infective stage, when incubated at a higher temperature. As regards high temperatures, 12 hours exposure to 43° C. proved lethal for eggs in all stages of development.



Fig. 32.15. Roundworms (*Ascaridia galli*) from small intestine of a chicken. (J. E. Ackert.)

**Pathology.** Ackert (1940) found that chickens infested with a large number of ascarids suffer from loss of blood, reduced blood sugar content, increased urates, shrunken thymus glands, retarded growth, and greatly increased mortality. Droopiness, emaciation, and diarrhea are the common clinical symptoms manifested by heavily parasitized birds (Fig. 32.17 A and B).

Experimental evidence is available to show that chickens three months or older manifest considerable resistance to infection with *Ascaridia galli*. Ackert, Edgar, and Frick (1939) reported that the increased number of goblet cells found in the epithelial lining of the duodenum of chickens at

three months or older may in some measure be responsible for the greater resistance to this nematode developed by these birds. The age at which the peak of the goblet cell formation occurred was found to correspond very closely to the development of the maximum resistance of the chickens to the growth of the nematodes.

Ackert and Beach (1933) showed that diets consisting chiefly of animal proteins and with little or no plant protein were important in aiding the



FIG. 32.16. *Ascaridia galli* ova. ×400. (E. A. Benbrook, 1928.)

chicken to build up resistance to infection with ascarids, and that diets consisting chiefly or wholly of vegetable proteins lowered the resistance to ascarid invasion. Alicata (1938), likewise, observed that birds given a diet consisting principally of animal protein concentrates developed fewer worms than those which were given a diet low in animal protein. Diets high in vitamins A and B (complex) have been shown to increase the fowl's resistance to *Ascaridia galli*, and diets low in these vitamins definitely favor parasitism.

Experiments conducted by Ackert, Eisenbrandt, Wilmoth, Glading, and Pratt (1935), which extended over a period of years and involved 1,351 chickens showed that the heavier breeds such as the Rhode Island Reds and

White and Barred Plymouth Rocks were more resistant to ascarid infections than the lighter White Leghorns and White Minorcas.

Large roundworms, similar in size and appearance to *Ascaridia galli*, occur in the small intestines of pigeons, guinea fowls, wild turkeys, and other game farm species. *Ascaridia numidae* of the guinea fowl, and *Ascaridia columbae* of the pigeon are shorter and somewhat thicker than *A. galli* of the chicken. The guinea fowl ascarid is the smallest of the three species.

*Ascaridia dissimilis* has been found only in the wild turkey of this country, but has been reported by Vigueras (1931) from the domestic turkey in Cuba. This ascarid is very similar in appearance to *A. galli* but is somewhat smaller. Shillinger (1942) reported *Ascaridia compar* as a parasite of the small intestine of the bobwhite quail in the United States.

The life history of all the above ascarids is probably similar to that of *Ascaridia galli*. Although the life histories of *Ascaridia columbae* and *A. dissimilis* have been shown by the writer to be direct, a detailed account of the hatching of the eggs, the period the larvae spend in the mucosa of the intestine, such as has been recorded for *Ascaridia galli* by Ackert, has



Fig. 32.17. A—chicken infested with large roundworms of intestine. B—chicken of same age free of roundworms. (Ackert and Herrick.)

not been worked out for these ascarids. The life history of *Ascaridia numidae* has not been experimentally demonstrated.

Birds heavily infested with either *Ascaridia dissimilis, A. columbae,* or *A. numidae* are probably affected in a somewhat similar manner as those heavily infested with *A. galli*.

### *Heterakis gallinae* (Gmelin, 1790)

**Synonyms.** *Ascaris gallinae* Gmelin, 1790; *Heterakis papillosa* Railliet, 1885, not *Ascaris papillosa* Bloch, 1782.

**Description.** Worms small, white. Head end bent dorsally. Mouth surrounded by 3 small, equally sized lips. Two narrow lateral membranes extend almost entire length of body. Esophagus ending in a well-developed bulb.

*Male* 7 mm. to 1.3 cm. long. Tail straight, ending in a subulate point; 2 large lateral bursal wings. Preanal sucker well developed, with strongly chitinized walls and small semicircular incision in posterior margin of wall of sucker. Twelve pairs of caudal papillae; 4 pairs distinctly postanal, 4 pairs of raylike papillae and two pairs of sessile papillae adanal, and 2 pairs of raylike papillae in vicinity of sucker (Fig. 32.18 A). Spicules dissimilar, the
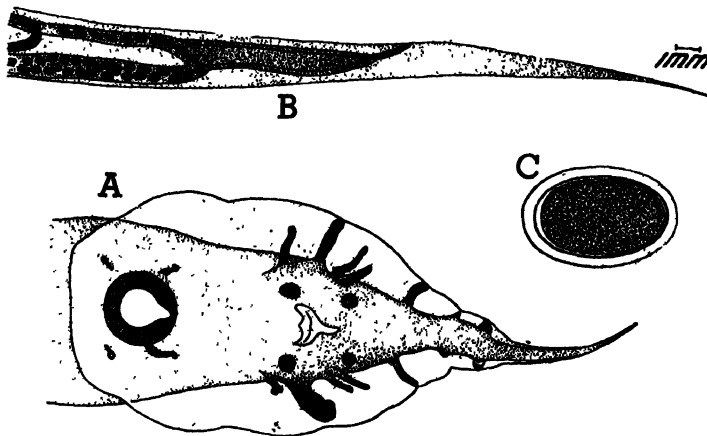


FIG. 32.18. *Heteiakis gallinae.* A—male tail. B—female tail. (After Lane, 1917.) C—egg. (After Cram, 1931.)

long one 2 to 2.17 mm. long, the short one 700μ to 1.1 mm. long.

*Female* 1 to 1.5 cm. long. Tail long, narrow, and pointed (Fig. 32.18 B). Vulva not prominent, slightly posterior to middle of body. Eggs thick-shelled, ellipsoidal, unsegmented when deposited (Fig. 32.18 C).

*H. gallinae* has been reported from the ceca of chickens, turkeys, guinea fowls, bobwhite quail, pheasants, and many other birds.

**Life history.** The eggs pass out in the feces in an unsegmented state. In approximately two weeks or less, under favorable conditions of temperature and moisture, these eggs will have reached the infective stage. When the latter are swallowed by a susceptible host, the embryos hatch from the eggs and develop to adult worms in the ceca. Roberts (1937) stated that the eggs hatched in the upper part of the intestine and at the end of 24 hours the majority of the young worms have reached the ceca. Aside from a short period in the cecal mucosa, 2 to 5 days, according to Uribe (1922), the entire life of the cecal worm is spent in the lumen of the cecum. At necropsy, the majority of the adult worms are found in the tips or blind ends of the ceca.

Earthworms may ingest the eggs of the cecal worm and may be the means of causing an infection in poultry, as the latter are very fond of earthworms.

**Pathology.** The pathogenicity of this species of roundworm has not been adequately studied by helminthologists. Riley and James (1922) observed that the ceca of experimentally infested birds showed marked inflammation and thickening of the walls.

The chief economic importance of the cecal worm lies in its role as a carrier of the blackhead organism, *Histomonas meleagridis.* Graybill and Smith (1920) demonstrated by experimental methods that blackhead may be produced in susceptible birds by feeding embryonated eggs of *Heterakis gallinae* taken from blackhead infected birds. These authors were of the opinion that the cecal worms lowered the resistance of the host to such a degree that the protozoan parasites already present were able to multiply to disease-producing proportions. Tyzzer (1926) presented evidence which indicated that the protozoan parasite is incorporated in the worm egg; however, he was unable to demonstrate the presence of the protozoan parasite within the egg.

Two other species of cecal worms, *Heterakis beramporia* and *Heterakis isolonche,* occur in the ceca of chickens and pheasants, respectively. So far as is known, the former species does not occur in poultry of the United States. However, the latter species has been found in pheasants in Pennsylvania and Connecticut. Both of these heterakids produce nodules in their respective hosts.

### Subulura brumpti

*Subulura brumpti* has been reported by Alicata (1940) to be a common pinworm of chickens in the Hawaiian Islands. Cram (1926b) and Dikmans (1929) reported this pinworm as occurring in the turkey in Puerto Rico. Foster (1939) collected it from the fowl in Panama, and Ward (1945) listed it as a parasite of the quail in Mississippi (Fig. 32.19 A and B).

No evidence that the larvae penetrated the cecal wall of the bird host for any part of its development, nor that they produced any extensive inflammatory tissue reaction was reported by Alicata (1940).
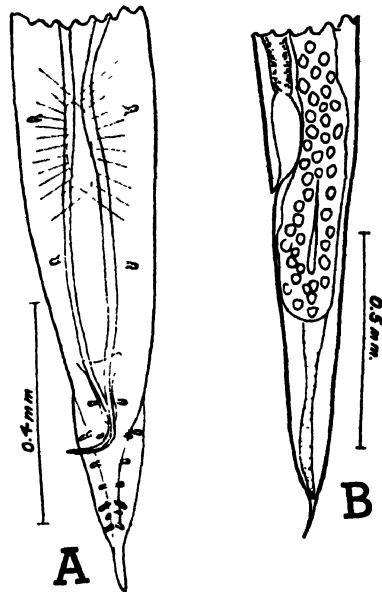


Fig. 32.19. *Subulura brumpti.* A—posterior end of male, ventral view. B—posterior end of female, lateral view. (From Cuckler and Alicata, 1944.)

Another cecal worm, *Subulura strongylina,* has been reported by Venard (1933) from the bobwhite quail in Ohio; by Cram (1927) from the chicken and guinea fowl in Puerto Rico; by Dikmans (1929) from the guinea fowl and by Van Valkenberg (1938) from poultry in Puerto Rico. Various insects, such as beetles and earwigs, serve experimentally as intermediate hosts of this cecal worm.

### TRICHURIDAE

For family diagnosis, see page 770.

<p style="text-align:center;">*Capillaria columbae* (Rudolphi, 1819)</p>

**Synonyms.** *Trichosoma columbae* Rudolphi, 1819; *Capillaria dujardini* Travassos, 1914.

**Description.** Worms hairlike.

*Male* 8.4 mm. to 1.2 cm. long by 49 to 53μ wide. Cloacal aperture almost terminal, with a small bursal lobe on either side, the two lobes connected dorsally by a delicate bursal membrane (Fig. 32.20 A). Spicule sheath with transverse folds; spicule 1.1 to 1.58 mm. long.

*Female* 1 to 1.8 cm. long by approximately 80μ wide. Vulva on slight prominence, slightly posterior to union of esophagus and intestine (Fig. 32.20 C). Eggs slightly brownish, lemon-shaped, thick-shelled.

This hairworm occurs in the small intestine of the domestic and wild pigeon, chicken, and turkey in the United States.

Madsen (1945) believes that *Capillaria dujardini* Travassos, 1915, and not *Capillaria columbae* Rudolphi, 1819, is the species found in the small intestine of the pigeon, chicken, partridge, and pheasant of Denmark. Furthermore, he feels that *Capillaria columbae* of Graybill, 1924, belongs to a new species which he designated as *Capillaria obsignata.* Inasmuch as the capillarids of birds in the North American continent need to be studied more critically from a taxonomic standpoint, the specific names now in vogue for those species of threadworms occurring in poultry in this country shall be used until such study may indicate that certain changes should be made.

**Life history.** *Capillaria columbae* has a direct development. The freshly deposited eggs are unsegmented and require from 6 to 8 days to develop completely formed embryos. The embryos do not escape from the eggs until after they have been swallowed by a susceptible host. The larvae enter the mucosa of the duodenum and apparently complete their development there. A few sexually mature adults were removed by the writer from the small intestine of a pigeon necropsied 19 days after the ingestion of the embryonated eggs, and a large number of similarly developed adults were removed from the small intestine of a pigeon necropsied 26 days after infection. Fecal

examination of the latter pigeon at the time of necropsy showed the presence of eggs.

It has been experimentally demonstrated that pigeons, when once infested with *Capillaria columbae* and held under conditions designed to preclude reinfection, will remain infested for about nine months.

*Capillaria columbae* has occasionally been reported from the small intestines of chickens and turkeys raised under natural conditions. Graybill (1924) stated that as a result of many necropsies of chickens and turkeys, this roundworm was never observed in these birds in large numbers. Wehr (1939) was successful in obtaining natural infestations in chickens and turkeys, but no heavy infestations were encountered in the latter.

**Pathology.** Birds heavily infested with *Capillaria columbae* spend much of their time apart from the rest of the flock, huddled on the ground, underneath the roosts, or in some corner of the room. Such birds show definite symptoms of emaciation and diarrhea. The feathers around the vent frequently appear ruffled and soiled, and the skin and visible mucous membranes are more or less pale. Death is often the result of heavy infestations. Levine (1938) reported that the first clinical symptoms of infestation in chickens appeared on the twelfth day after experimental inoculation of embryonated eggs. At this time the feces contained much pinkish material composed of mucus, necrosed epithelial cells, and numerous erythrocytes, granulocytes, and lymphocytes. From the twelfth to the sixteenth days the



Fig. 32.20. *Capillaria columbae.* A—ventral view, and B—lateral view, of male tail. (After Graybill, 1924.) C—region of vulva. (After Eberth, 1863, slightly modified.)

feces of the birds were watery and contained large quantities of epithelium and inflammatory exudate which was being eliminated from the intestinal tract. Following this period, most of the infested birds regained their normal appearance, and the feces became normal. However, many of the birds lost weight steadily, became extremely emaciated, and either died or were

destroyed because of a weakened condition. In fatal and in advanced cases of infestation, the intestines showed extensive destruction of the mucosa, often with complete sloughing of the mucous membrane. The intestines usually contain a large quantity of fluid. In nonfatal experimental cases the intestinal wall was thickened considerably owing to the edematous infiltration.

Another threadworm, *Capillaria longicollis* [=*C. caudinflata* (Molin, 1858) and *C. meleagris-gallopavo* (Barile, 1912)], has been occasionally found in chickens, turkeys, and pheasants in the United States. This species of capillarid worm may be differentiated easily from other species of the same genus found in poultry of this country by the presence on the male tail of two large lateral transparent membranes just anterior to the cloacal aperture, and on the female of a membranous tubular or trumpet-shaped projection in the region of the vulva.

Barile (1912) found these worms in turkeys showing hemorrhagic, croupous enteritis, but was unable to state positively whether the worms were responsible for this pathological condition. Baker (1930), in connection with frequent findings of this worm in the province of Quebec, Canada, noted that the worms were associated with ulcerous patches varying in size from pin-point areas to greatly extended and hardened areas. An infested chicken observed by Graham, Thorp, and Hectorne (1929) in Illinois showed weakness, anemia, and emaciation before death. At necropsy, the intestine just anterior to the ceca was markedly dilated, with a follicular diphtheritic enteritis present.

### TRICHOSTRONGYLIDAE

For family diagnosis, see page 783.

#### *Ornithostrongylus quadriradiatus* (Stevenson, 1904)

**Synonym.** *Strongylus quadriradiatus* Stevenson, 1904.

**Description.** Worms delicate, slender, red when freshly collected, apparently from ingested blood in intestine. Cuticle about head inflated to form vesicular enlargement (Fig. 32.21 A).

*Male* 9 mm. to 1.2 cm. long. Bursa bilobed, with no distinct dorsal lobe. Dorsal ray much shorter than other rays, not extending halfway to bursal margin, bifurcating near its tip to form 2 short tips, and a stumpy process present on each side near base of ray. Spicules equal, 150 to 160μ long, somewhat curved, each terminating in 3 pointed processes (Fig. 32.21 B). Telamon 57 to 70μ long, with two longitudinal processes extending backward and forward along dorsal wall of cloaca, and two lateral processes forming a partial ring through which the spicules protrude.

*Female* 1.8 to 2.4 cm. long. Vulva near end of tail. Vagina short, followed

by 2 powerful muscular ovejectors. Tail tapers to a narrow, blunt end, bearing a short spine. Eggs segmenting when deposited.

This bloodsucking nematode occurs in the small intestine of pigeons, turtle doves, and mourning doves in the United States.

**Life history.** The oval, thin-shelled eggs are voided in the droppings and hatch in approximately 19 to 24 hours under favorable conditions of moisture and temperature. After escaping from the egg, the young larva molts twice within the next 3 or 4 days. It has now reached the infective stage.
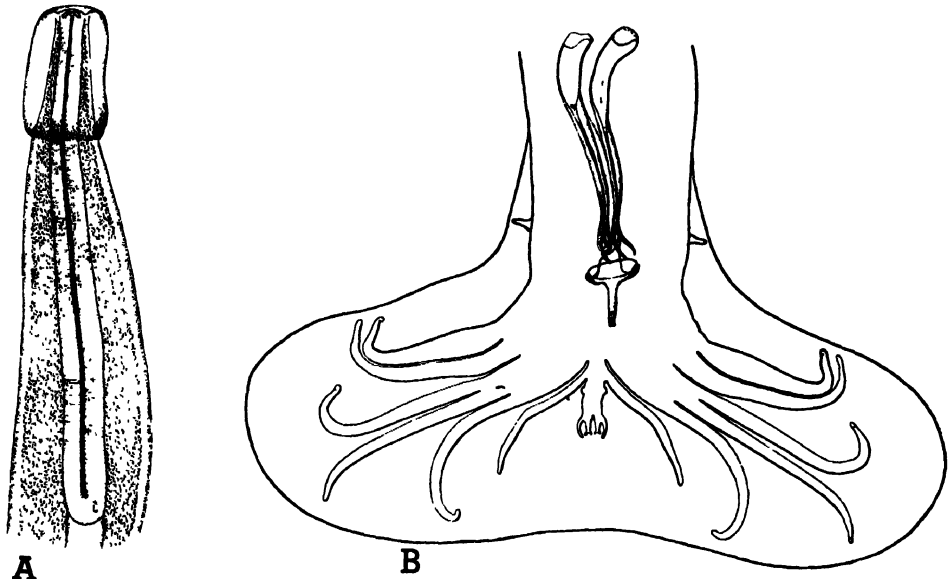


Fɪɢ. 32.21. *Ornithostrongylus quadiiradiatus.* A—anterior end. B—caudal bursa of male. (After Stevenson, 1904.)

When the infective larva is swallowed by a pigeon or other susceptible host, it grows to maturity in the small intestine. The female worm begins to deposit eggs in 5 or 6 days following ingestion of the larva.

**Pathology.** Stevenson (1904) observed that this parasite was the cause of many deaths among a flock of fancy pigeons in Washington, D. C. Le Roux (1926, 1930) mentioned this roundworm as having caused serious losses in a flock of valuable imported pigeons. Vigueras (1929) reported similar losses among pigeons in Cuba, and Kamarov and Beaudette (1931) attributed large numbers of deaths among squabs as having been due to this bloodsucking parasite. These investigators are agreed that deaths among the birds were attributable principally to a catarrhal enteritis and a loss of blood due to hemorrhage.

Birds heavily infested with *Ornithostrongylus quadriradiatus* behave

much the same as birds heavily parasitized with other bloodsucking parasites. They become droopy, remain squatted on the ground or floor, and if disturbed, they try to move but usually tip forward on the breast and head. Food is eaten sparingly and is frequently regurgitated, along with bile-stained fluid. There is a pronounced greenish diarrhea, and the bird gradually wastes away. Symptoms of difficult and rapid breathing usually precede death. The intestines of fatally infested birds are markedly hemorrhagic and have a greenish mucoid content, with masses of sloughed epithelium (Fig. 32.22).



Fig. 32.22. Section of duodenum of pigeon during later stage of infestation with *Ornithostrongylus quadriradiatus*, showing: a—sloughing of mucosa; b—necrotic areas; and c—lymphocytic infiltration. ×150. (After Cuvillier, 1937.)

### *Trichostrongylus tenuis* (Mehlis, 1846)

**Synonyms.** *Strongylus tenuis* Mehlis, 1846 (in Creplin, 1846) ; *Strongylus pergracilis* Cobbold, 1873; *Trichostrongylus pergracilis* (Cobbold, 1873) Railliet and Henry, 1909.

**Description.** Worms small and slender. Body gradually attenuated in front of genital opening. Mouth surrounded by 3 small, inconspicuous lips. Cuticle of anterior end of body lacking conspicuous striations for a distance of about 200 to 250µ from extremity, then with distinct serrated appearance for a distance of about 1 to 2 mm. more.

*Male* 5.5 to 9 mm. long by 48µ wide near center of body. Cuticle inflated on ventral surface just anterior to bursa. Two lateral and one dorsal lobe of bursa, the dorsal lobe not distinctly marked off from lateral lobes. Each

lateral lobe supported by 6 rays (Fig. 32.23 A). The dorsal ray bifid at its distal third, and each of these divisions again bifid and very finely pointed (Fig. 32.23 B). Spicules dark brown in color, slightly unequal in length, the longest 120 to 164μ long, the shortest 104 to 150μ long; both much twisted, especially at distal ends, and provided with an earlike structure on proximal end (Fig. 32.23 C). Both spicules apparently surrounded in distal two-thirds by a thin membrane extending for a short distance beyond distal ends. Gubernaculum strongly cuticularized along margins, spindle-shaped in ventral and dorsal views (Fig. 32.23 D and E).

*Female* 6.5 mm. to 1.1 cm. long by 77 to 100μ wide at level of vulva. Vulva in posterior part of body, with crenulated edges. Uteri divergent. Eggs thin-shelled.

It was concluded by Cram and Wehr (1934), as the result of a critical study of a large number of specimens of the génus Trichostrongylus collected from the ceca of both American and European domestic and wild game birds, that the material thus examined represented only one species, instead of two, as previously suspected. Although *Trichostrongylus pergracilis* had been described from the ceca of many birds, the differences between the description of this species and that of *T. tenuis* were inconsequential and not of specific value. As a result of the above study, therefore, the authors concluded that *T. pergracilis* and *T. tenuis* were identical



FIG. 32.23. *Trichostrongylus tenuis.* (After Cram and Wehr, 1934.) A—bursa, lateral view; semidiagrammatic. B — dorsal and externo-dorsal rays of bursa, showing variation which may occur in length of latter. C—right and left spicule, ventral view. D—gubernaculum, lateral view. E—gubernaculum, dorsal view. A and B from European partridge, C to E from red grouse. Scale refers to camera lucida drawings, B to E, inclusive.

morphologically, and since the specific name *tenuis* had priority over *pergracilis*, the former name was accepted as the valid name.

In the United States, *T. tenuis* has been collected from the pheasant, *Phasianus colchicus;* the blue goose, *Chen caerulescens;* the Canadian goose, *Branta canadensis;* the domestic goose, *Anser anser domesticus;* the guinea

fowl, *Numida meleagridis;* the chicken, *Gallus domesticus;* the turkey, *Meleagris gallopavo;* and the bobwhite quail, *Colinus virginianus.* The red grouse, *Lagopus scoticus,* and the European partridge, *Perdix perdix,* are the only two hosts from which *T. tenuis* has been collected in Europe. According to Cram (1931a) this trichostrongyle occurs widely among the quail of the southeastern United States.

**Life history.** This worm has a direct life history. The eggs hatch within 36 to 48 hours after they have been passed in the droppings of the infested bird. The larvae become infective within approximately two weeks following expulsion of the eggs in the droppings. Within this time the larvae have molted twice. When the latter are picked up by a susceptible host, the infective larva molts twice more within the ceca of the bird before finally becoming an adult.

*Trichostrongylus tenuis* from pheasants has been successfully transmitted to the domestic turkey, guinea fowl, and chicken.

**Pathology.** *T. tenuis* produces definite clinical symptoms when present in large numbers. The changes in the ceca consist of a thickening and a reddening of the walls, and small hemorrhages are sometimes present. Loss of weight, anemia, and chronic toxemia have been reported as symptoms resulting from heavy infections.

### STRONGYLOIDIDAE

Members of this family are characterized by having an alternation of generations, the free-living generation consisting of males and females while the parasitic generation consists of hermaphroditic females only .

### *Strongyloides avium* (Cram, 1929)

**Description.** Parasitic generation, consisting of parthenogenetic females only, in intestine of avian host, and free-living generation, consisting of both males and females, in soil.

*Parasitic adult.* 2.2 mm. long by 40 to 45µ wide. Vulva with projecting lips, located 1.4 mm. from head end. Uteri divergent from vulva; ovaries recurrent in simple "hair-pin bends," their course not sinuous. Eggs with very thin shells, segmenting when deposited.

Cram (1929) reported the occurrence of this extremely small roundworm from the ceca of chickens in Louisiana. Later (1936b), this same author reported this species from the ceca and small intestines of chickens in Puerto Rico. The junco, *Junco hyemalis hyemalis,* in Virginia, and the coot, *Fulica americana,* in North Carolina have been found to harbor natural infestations of this parasite.

**Life history.** The eggs hatch soon after being passed in the droppings, sometimes as soon as 18 hours. The young worms develop in the soil to adult males and females. Shortly thereafter the females give rise to young,

which feed, molt, and develop into other adult free-living males and females, or they may transform into another type of larvae known as the infective larvae. When these infective larvae are swallowed by a susceptible host, infestation results. Unlike most species of nematodes, the parasitic cycle of *Strongyloides avium* consists of females only, no parasitic males having been found.

**Pathology.** During the early or acute stage of the infestation the walls of the ceca are greatly thickened; typical pasty cecal contents almost disappear, the discharge being thin and bloody. If the fowl survives this acute stage, the ceca gradually become functional again, and the thickening of the walls decreases. Young birds suffer most from infestations with this worm. If the infestation is light or if the birds are adults, little, if any, clinical effect has been noted.

### CONTROL OF POULTRY NEMATODES

**Prevention.** Preventive measures for the control of poultry roundworms have been developed along the lines of sanitation, hygiene, and management, and it is along these lines that the greatest progress has been made.

The proper selection of a permanent site for the poultry runs is one of the first essentials to the maintenance of health among the birds. The land should be sloping and of a sandy or gravelly nature to provide for proper drainage. If the soil is heavy, or the lay of the land is such as to render natural drainage impossible, artificial drainage should be provided. The nonparasitic stages of helminth parasites require moisture for their proper development. Therefore, the presence of surface water, which birds are apt to drink, must be regarded as unhealthful, and provisions should be made to eliminate it as soon as possible. Damp places and water holes are ideal breeding places for many of the intermediate hosts of poultry nematodes.

The practice of rotating the birds from one area of land to another in order to reduce parasitism among the birds has been followed with reasonably good success in some sections of the country. The four-yard system is the one most widely advocated and probably the one best suited for general use. A given area of land—the amount depending upon the number of birds raised—is cross-fenced so that it is divided into four equal lots. The shelter or house is placed in the center of the plot and so constructed that a door opens into each pen. The birds are rotated from one pen to another, keeping them in each pen not longer than two or three months. After removing the birds from one of the pens, the ground in that pen may be prepared for planting to some green crop or left undisturbed to undergo self-sterilization by exposing the infected droppings to the direct action of the sun, wind, and cold. The planting of the yards to a permanent or temporary crop serves a twofold purpose: (1) It furnishes abundant green food for the growing birds,

and (2) there is some evidence to indicate that birds are less likely to pick up contaminated soil when plenty of green food is available, thus reducing parasitism in the birds. The house and adjacent grounds should be cleaned as often as is deemed necessary to maintain good sanitation. The practice of removing the soil about the house to a depth of 6 to 8 inches and replacing it with sand or coarse ashes has sometimes been followed. During the summer months, birds spend a great deal of their time in the shade of the house and it is necessary that extra precautions be taken to improve the sanitary conditions around the house.

The frequent removal of the droppings and the proper disposal of them cannot be recommended too highly. (See discussion of manure disposal under control of cestodes, p. 835.)

The raising of the different species of birds together or in close proximity to each other is a dangerous procedure as regards parasitism. Ransom (1921) showed by careful investigations that adult turkeys served as carriers of gapeworms in transmitting gapeworm disease to little chicks and that the older chickens were almost entirely insusceptible to infection with the above worms. Adult turkeys carrying natural infestations of gapeworms apparently suffer only slightly, while young chicks and turkey poults suffer very severely from infestations with gapeworms.

It is also dangerous for turkeys to associate with chickens. Tyzzer (1928) established the fact that blackhead occurs in young and old chickens, the latter usually recovering from the disease without suffering a great deal. However, this same investigator also demonstrated that the recovered chickens remained carriers of the blackhead organism for an indefinite period, and that turkeys contract blackhead by exposure to infected poultry or runs occupied by the latter.

It follows from the work of Ransom, Tyzzer, and others that anyone wishing to raise poultry would do well to decide in the beginning to raise turkeys or to raise chickens but not to raise both on the same land or in close proximity to each other. Should a person desire to raise both chickens and turkeys, he should keep the two kinds of birds on ranges well separated from one another.

It is dangerous too for turkeys and chickens to associate with guinea fowls. Wehr (1939b) showed experimentally that the guinea fowl is susceptible to infection with the poultry gapeworm, *Syngamus trachea,* at any age and that this bird may carry the parasites for as long as 98 days.

**Treatment.** Satisfactory remedial agents have been discovered for the removal of the poultry gapeworm, *Syngamus trachea,* the large intestinal roundworm of the chicken, *Ascaridia galli,* and the cecal worm, *Heterakis gallinae,* but wholly effective drugs for the removal of the other poultry nematodes are lacking.

Wehr, Harwood, and Shaffer (1939) found that the compound, barium antimonyl tartrate, successfully removed a very high percentage of gapeworms from chicks, turkey poults, and adult turkeys when administered as an inhalant.

For treatment, the birds are placed in a suitable container, such as a tight box. The drug is introduced into the box as a very fine powder by means of a dust gun. Because of its fluffiness, the powder remains suspended in the air for a long time. As the dust-laden air is breathed in by the infested birds, the fine particles of barium antimonyl tartrate apparently adhere to the moist surfaces of the worms and act as a contact poison.

The size of the dose of the powder to be administered is determined by the cubic capacity of the treatment box. One ounce of barium antimonyl tartrate has been found to be sufficient for a box having a capacity of 8 cubic feet. Approximately one-third of the powder is introduced into the treatment box at the first operation. If the box is of a convenient size to lift, it is tilted slowly from side to side several times. Tilting the box causes the birds to flap their wings, thus aiding in redispersing any powder that may have settled on the feathers of the birds or on the floor of the box. Tilting also causes the birds to breathe deeper and more frequently. Deep breathing is necessary to bring the powder in contact with the worms that may be found in the lower part of the trachea. In the case of mature birds, when the treatment box is likely to be too large and heavy to tilt by hand, a small electric fan may be placed on the floor of the box to keep the powder agitated during the process of treatment. After about 5 minutes, one-half of the remaining powder is blown into the box and the tilting or the use of the fan is repeated. The remaining one-third of the powder is introduced into the treatment box about 15 minutes following the introduction of the first one-third, and the box is again tilted or the fan used. The birds are released 5 to 10 minutes after the last of the powder has been blown into the box.

Hall and Shillinger (1923a) reported that carbon tetrachloride failed to remove any large roundworms from one chicken treated with 1 cc. per kilo of body weight, but removed all the worms present in three cases when administered at the rate of 2, 4, and 5 cc. per kilo of body weight. Ackert and Graham (1935) found that carbon tetrachloride was highly efficacious in removing the large intestinal roundworm from young chickens at a dose rate of 4 cc. per kilo with apparently no ill effects.

Rectal injections of a mixture of oil of chenopodium and of olive oil or cottonseed oil, given with a hard rubber syringe, in doses of 0.1 cc. in 5 cc. of the oil in case of birds weighing 1.5 pounds and double this amount of chenopodium and oil for adult birds weighing 3 pounds or over, have been found by Hall and Shillinger (1923b) to be approximately 90 per cent efficacious for the removal of the cecal worm of chickens and turkeys.

McCulloch and Nicholson (1940) reported that phenothiazine when given either in repeated or single doses was very effective for the removal of the cecal worm from chickens. Doses ranging between .05 and 0.5 grams were found to be the most satisfactory individual doses. These authors found that repeated doses and the administration of the drug in individual capsules to be slightly more satisfactory, although they indicated that flock medication appeared to be practical. They experimentally determined that doses up to 500 times the smallest amount found to be therapeutically effective, had no apparent harmful effect on the birds; such massive doses also had no anti-heterakid effect. The administration of phenothiazine in doses of 0.5 grams per bird had no appreciable effect on a flock in egg production and was not followed by enteritis or other digestive disturbances, except for a slight softening of the feces 24 hours after treatment.

It has been known for many years that nicotine possessed a high nematodicidal action against the large roundworm of poultry. However, because of its toxicity to the fowl, its use for the control of this parasite was temporarily delayed.

Herms and Beach (1916) were the first to employ tobacco stems for the removal of poultry roundworms. They found that by soaking chopped tobacco stems for 2 hours and mixing the stems and the liquid with about one-third of their normal ration of mash, many roundworms were removed. The birds were fasted for about 24 hours before they were given the medicated mash.

Freeborn (1923), of the University of California Agricultural Experiment Station, conceived the idea of mixing nicotine sulphate (Black Leaf "40") with an earthen material known as Lloyd's alkaloidal reagent, thereby rendering much less toxic a substance which by itself would not be safe to use. The mixture, which contained 6.6 cubic centimeters of nicotine sulphate and 16 grams of Lloyd's alkaloidal reagent, was placed in capsules, each capsule containing approximately 350 to 400 milligrams, and administered individually to the birds. This formula became known as the University capsule.

The introduction of this capsule marked a renewed interest in the treatment of fowls for the removal of the large intestinal roundworm. However, it was soon discovered that the gelatine capsule containing the mixture was soluble in the secretions of the upper part of the digestive tract and toxic symptoms and, in some cases, death resulted. Moreover, autopsies on some of the treated birds revealed the presence of numerous roundworms, indicating that the dose of nicotine in the University capsule was not sufficient to remove all the worms satisfactorily.

Davis (1940) stated that it was possible to mix nicotine with an organic colloidal material (name not given) to obtain a mixture which was non-

lethal to the animal to which it is administered even though the amount of nicotine is in excess of a lethal dose. The use of such a mixture, he stated, satisfactorily removed roundworms from chickens, provided 70 to 80 milligrams of nicotine was administered at a single dose. He further stated that it was possible in the mixing of the nicotine and the organic colloidal material to so regulate the release of the alkaloid (nicotine) that the greatest amount could be liberated where it was most needed. In case of the intestinal roundworm, which is found to be most numerous in the anterior portion of the small intestine, the liberation of the drug would have to be delayed until after it passed through the gizzard.

Levine (1936) administered a mixture containing one-fourth pound Black Leaf worm powder (a 5 per cent nicotine compound composed of nicotine sulphate mixed with a special fuller's earth) and 5 pounds of mash to forty-five pullets. Feed was withheld from the birds overnight. The birds promptly cleaned up the mash the next morning. Two hundred and eighty-four worms were removed by the treatment; none was found at autopsy.

Guthrie and Harwood (1942) conceived the idea of mixing phenothiazine and nicotine-bentonite (a claylike material) and the mixture administered for the removal of both *Heterakis gallinae* and *Ascaridia galli* from chickens. Tablets containing 33 parts phenothiazine, 66 parts nicotine-bentonite (5 per cent nicotine), and 1 part sodium stearate removed 83.7 per cent of 1,012 Heterakis and 96.2 per cent of 131 Ascaridia. When administered separately and in equivalent amounts to infested chickens, the phenothiazine removed 89.9 per cent of 675 Heterakis and 48.2 per cent of 110 Ascaridia; the nicotine-bentonite removed 10.1 per cent of 1,246 Heterakis and 85.2 per cent of 149 Ascaridia.

Harwood and Stuntz (1945) found that phenothiazine and nicotine-bentonite mixture gave good results in removing *Heterakis gallinae* and only fair results in removing *Ascaridia dissimilis* from the turkey.

Scientists of the Bureau of Animal Industry demonstrated that the feeding of a medicated mash containing 15 grams of nicotine sulphate (Black Leaf "40"), 151 grams of phenothiazine, 287 grams of bentonite, and 44 pounds of chick feed for 3 consecutive days at intervals of three weeks to chickens held continuously on worm-infested soil maintained a low level of parasitism in the treated birds.

For the treatment of the eyeworm, Sanders (1929) recommended a modification of that advocated by Wilcox and McClelland in 1913. The eye is first anesthetized by means of a local anesthetic. The worms are exposed by lifting up the nictitating membrane and one or two drops of a 5 per cent solution of creolin is placed directly on the worms. The eye is then immediately irrigated with pure water to remove the excess creolin solution. Inasmuch as the worms are killed immediately upon contact with the creolin

solution, the irrigating of the eye does not interfere with the effectiveness of the treatment. Within 48 to 60 hours after the treatment, the eyes will begin to show improvement, provided the damage has not been too great.

Emmel (1939) reported that the feeding of regular mash to which
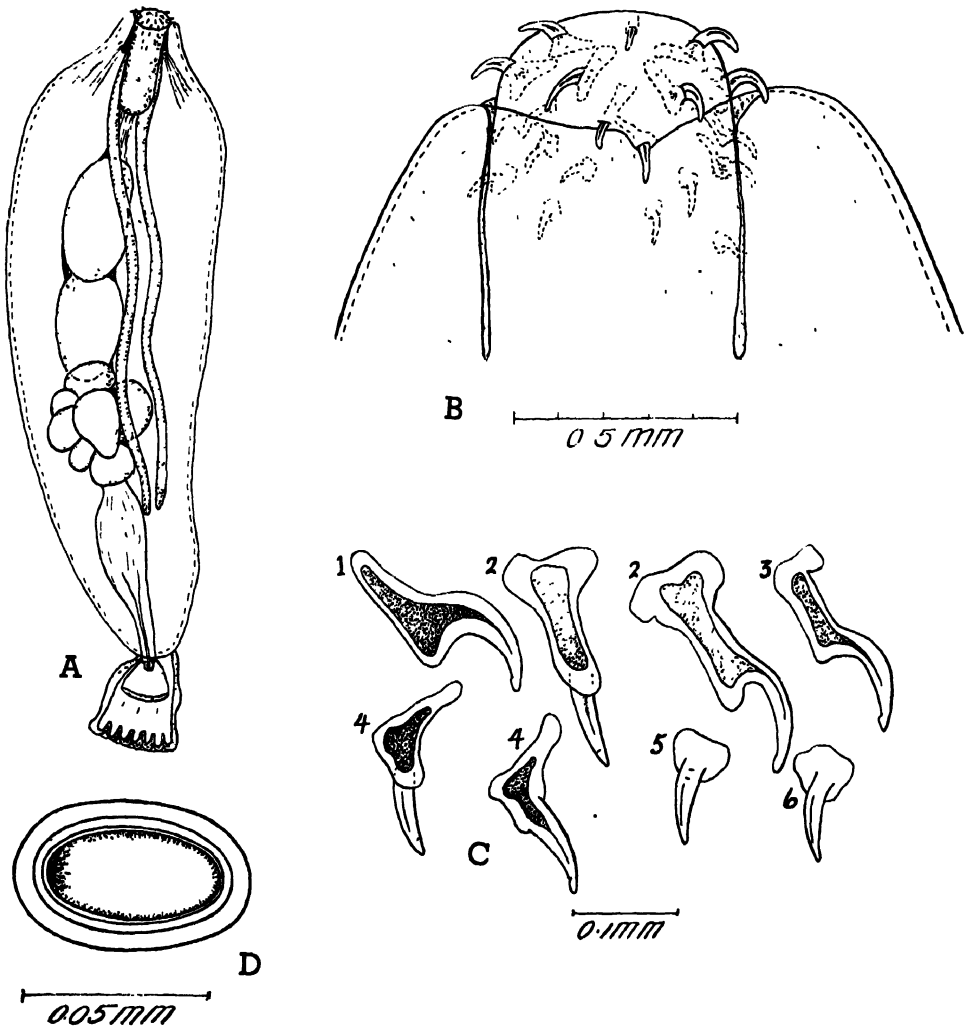


Fig. 32.24. *Oncicola canis.* A—male showing reproductive organs. B—proboscis. C—hooks from proboscis (numerals indicate row). D—egg. (From Price, 1926.)

5 per cent of flowers of sulphur had been added seemed to benefit turkeys infected with *Capillaria contorta.* At the end of three weeks' treatment, he stated that "recovery occurred in all affected birds which were able to eat when treatment was started."

## ACANTHOCEPHALA

The Acanthocephala or thorny-headed worms are parasites occurring as adults in the intestinal tract of vertebrates. In form they are elongate, roughly cylindrical, or spindle-shaped. Several distinct body regions are
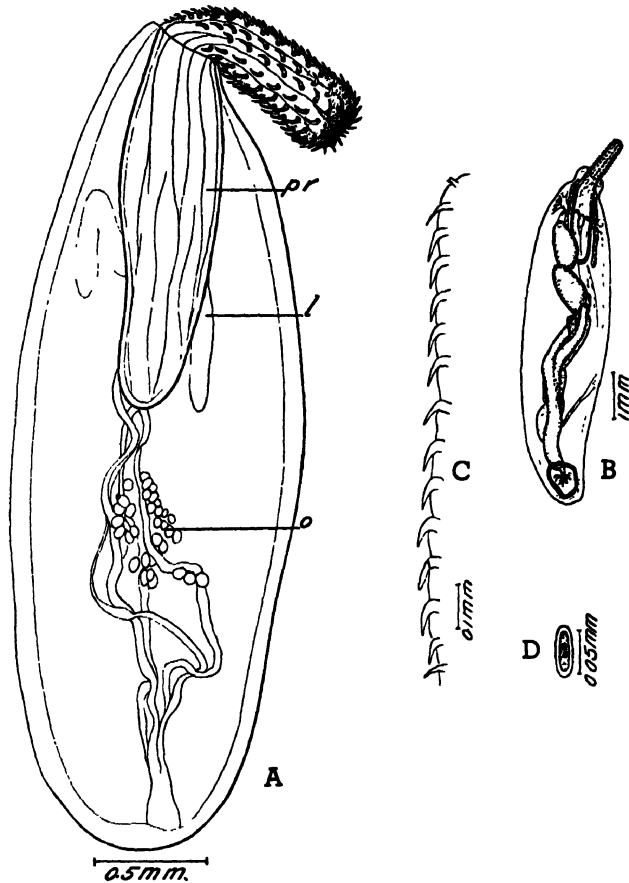


FIG. 32.25. *Plagiorhynchus formosus*. A—young female (enlarged): 1, lemniscus; o, ovary; pr, proboscis receptacle (from Jones, 1928). B—male. C—hooks from proboscis. D—egg. (B, C, D, enlarged.) (From Van Cleave, 1918.)

recognizable: retractile proboscis armed with hooks, a neck, and a body proper. The retractile proboscis bears always a considerable number of recurved hooks which are arranged in rows. The number, form, and arrangement of the hooks are valuable diagnostic characters. The body proper forms the major portion of the worm. It is usually unarmed but may bear small spines of definite form and arrangement on some portion of the external surface. This group of worms is deprived of a digestive tract.

Nutrition is thus provided for entirely by absorption through the body wall. The sexes are separate in all cases. The male is smaller and more slender than the female and often distinguished externally by a bell-shaped bursa that surrounds the genital pore.

So far as known, all species of Acanthocephala require one or more intermediate hosts before reaching a stage of development where they are infective for the final host. Various arthropods, snakes, lizards, and amphibians serve as hosts of the larval stages of these parasites.

Only three species of thorny-headed worms have been reported as parasites of domestic poultry in North America, two of these as immature forms.

### Oncicola canis

Oncicola canis (Kaupp, 1909) was found in about 10 per cent of the young turkeys around San Angelo, Texas, by Price (1929). The worms were encysted under the epithelial lining of the esophagus in numbers varying from a few to 100 or more. They were reported as the possible cause of death (Fig. 32.24 A, B, C, and D).

The adults of this parasite occur in the dog and coyote. The presence of larval forms of this parasite in young turkeys suggests that such occurrences are accidental, the young



FIG. 32.26. Polymorphus boschadis. A—male. B—larva from Gamarus pulex. C—proboscis of larva. (Enlarged.) (From Lühe, 1911.)

worms encysting when taken into an unsuitable host.

### Plagiorhynchus formosus

An immature male and two female specimens of Plagiorhynchus formosus Van Cleave, 1918, were reported by Jones (1928) from the small intestine of a chicken collected at Vineland, New Jersey. Other bird hosts from which this species has been reported are the flicker, collected at Bowie, Maryland; the crow, collected at Washington, D. C.; and the robin in New Jersey (Fig. 32.25 A, B, C, and D).

### Polymorphus boschadis

Wickware (1922) reported Polymorphus boschadis Schrank, 1788, from

the duck in Canada. This parasite is reported as causing serious injury and death among domesticated waterfowl, especially in young birds. It causes an inflammation of the intestine with subsequent anemia and cachexia. According to Schlegel (1921), the birds are sick only a short time, the gait is staggering, and the head and wings droop (Fig. 32.26 A, B, and C).

## REFERENCES

Ackert, J. E.: 1931. The morphology and life history of the fowl nematode *Ascaridia lineata* (Schneider). Parasitology 23:360.

———: 1940. The large roundworm of chickens. Vet. Med. 35:106.

——— and Beach, T. D.: 1933. Resistance of chickens to the nematode, *Ascaridia lineata*, affected by dietary supplements. Trans. Am. Micr. Soc. 52:51.

———, Edgar, S. A., and Frick, L. P.: 1939. Goblet cells and age resistance of animals to parasitism. Trans. Am. Micr. Soc. 58:81.

———, Eisenbrandt, L. L., Wilmoth, J. H., Glading, B., and Pratt, I.: 1935. Comparative resistance of five breeds of chickens to the nematode *Ascaridia lineata* (Schneider). Jour. Agr. Res. 50:607.

——— and Graham, G. L.: 1935. The efficacy of carbon tetrachloride in roundworm control. Poultry Sci. 14:228.

Alicata, J. E.: 1938. Studies on poultry parasites. Rep. Hawaii Agr. Exper. Sta. (1937) :93.

———: 1940. Poultry parasites. Rep. Hawaii Agr. Exper. Sta. (1939) :66.

Allen, A. A.: 1925. The grouse disease in 1924. Bul. Am. Game Protect. Assn. 14:11, 12, 20.

——— and Gross, A. O.: 1926. Report of the ruffed grouse investigations; season of 1925–26. Am. Game 15:81, 86.

Baker, A. D.: 1930. The internal parasites of poultry in Quebec. Scient. Agr. 11:150.

Barber, L. B.: 1916. Live stock disease investigations. Ann. Rep., Guam Agr. Exper. Sta. (1915) :25.

Barile, C.: 1912. Sur une espèce de trichosome signalée chez le dindon *(Meleagris gallopavo domestica* (L.)). Bul. Soc. zool. (France) 37:126.

Bump, G.: 1935. Ruffed grouse in New York State during the period of maximum abundance. Trans. 21 Am. Game Conf.:364.

Bunyea, H., and Creech, G. T.: 1926. The pathological significance of gizzard-worm disease of geese. No. Am. Vet. 7:47.

Chitwood, B. G., and Chitwood, M. B.: 1937. An Introduction to Nematology. Baltimore. Section I, Part 1.

Clapham, P. A.: 1934. Experimental studies on the transmission of gapeworm *(Syngamus trachea)* by earthworms. Proc. Roy. Soc., London, series B, 115:18.

Cram, E. B.: 1926a. A parasitic nematode as the cause of losses among domestic geese. No. Am. Vet. 7:27.

———: 1926b. *Subulura brumpti* from the turkey in Puerto Rico. Jour. Parasit. 12:164.

———: 1926c. A parasitic disease of the esophagus of turkeys. No. Am. Vet. 7:46.

———: 1927. New records of distribution for various nematodes. Jour. Parasit. 14:70.

———: 1928. Nematodes of pathological significance found in some economically important birds in North America. U.S.D.A., Tech. Bul. 49:1.

———: 1929. A new roundworm parasite, *Strongyloides avium*, of the chicken, with observations on its life history and pathogenicity. No. Am. Vet. 10:27.

———: 1931a. Internal parasites and parasitic diseases of the bobwhite. Nematodes (roundworms) in quail. In Stoddard, H. L.: The Bobwhite Quail; Its Habits, Preservation, and Increase. Charles Scribner's Sons, New York, pp. 240–96.

———: 1931b. Developmental stages of some nematodes of the Spiruroidea parasitic in poultry and game birds. U.S.D.A., Tech. Bul. 227:1.

———: 1936a. Species of Capillaria parasitic in the upper digestive tract of birds. U.S.D.A., Tech. Bul. 516:1.

———: 1936b. Notes concerning internal parasites of poultry in Puerto Rico. Agr. Notes, Puerto Rico Agr. Exper. Sta., U.S.D.A., May 15, five mimeographed leaves.

——— and Wehr, E. E.: 1934. The status of species of Trichostrongylus of birds. Parasitology 26:335.

Crawford, M.: 1940. Infection of adult fowls with *Syngamus trachealis*. Indian Jour. Vet. Sci. and An. Husb. 10:293.

Cuckler, A. C., and Alicata, J. E.: 1944. The life history of *Subulura brumpti*, a cecal nematode of poultry in Hawaii. Trans. Am. Micr. Soc. 63:345.

Davis, D. E.: 1940. Nicotine in the control of *Ascaridia lineata* in fowls. Vet. Med. 35:109.

Dikmans, G.: 1929. Report of the parasitologist. Rep. Puerto Rico Agr. Exper. Sta. (1927) :27.

Emmel, M. W.: 1939. Observations on *Capillaria contorta* in turkeys. Jour. Am. Vet. Med. Assn. 94:612.

Foster, A. O.: 1939. Some helminthic parasites recovered from domesticated animals (excluding equines) in Panama. Proc. Helminth. Soc. Wash. 6:101.

Freeborn, S. B.: 1923. The control of the suckered roundworms of poultry. Cornell Vet. 13:223.

Goble, F. C., and Kutz, H. L.: 1945. The genus *Dispharynx* (Nematoda: Acuariidae) in galliform and passeriform birds. Jour. Parasit. 31:323.

Graham, R., Thorp, F., and Hectorne, R. L.: 1929. Capillaria in chickens. Jour. Am. Vet. Med. Assn. 74:1060.

Graybill, H. W.: 1924. *Capillaria columbae* (Rud.) from the chicken and turkey. Jour. Parasit. 10:205.

———. and Smith, T.: 1920. Production of fatal blackhead in turkeys by feeding embryonated eggs of *Heterakis papillosa*. Jour. Exper. Med. 31:647.

Guthrie, J. E., and Harwood, P. D.: 1942. The efficacy of phenothiazine and nicotine-bentonite for the removal of *Heterakis gallinae* and *Ascaridia galli* from chickens. Jour. Parasit. 28 (Suppl.) :24.

Hall, M. C., and Shillinger, J. E.: 1923a. Miscellaneous tests of carbon tetrachloride as an anthelmintic. Jour. Agr. Res. 23:163.

——— and Shillinger, J. E.: 1923b. The removal of heterakids from the ceca of chickens by rectal injections of anthelmintics. Jour. Am. Vet. Med. Assn. 62:623.

Harwood, P. D., and Stuntz, D. I.: 1945. Phenothiazine and nicotine-bentonite as an anthelmintic in turkeys. Proc. Helminth. Soc. Wash. 12:1.

Herms, W. B., and Beach, J. R.: 1916. Round worms in poultry—life history and control. Calif. Agr. Exper. Sta., Cir. 150.

Hung, S. L.: 1926. Pathological lesions caused by *Capillaria annulata*. No. Am. Vet. 7:19.

Itagaki, S.: 1927. On the life history of the chicken nematode, *Ascaridia perspicillum*. Rep. Proc. 3rd World's Poultry Cong., p. 339.

Jones, M.: 1928. An acanthocephalid, *Plagiorhynchus formosus*, from the chicken and robin. Jour. Agr. Res. 36:773.

Kaupp, B. F.: 1909. *Echinorhynchus canis*. Am. Vet. Rev. 35:154.

Komarov, A., and Beaudette, F. R.: 1931. *Ornithostrongylus quadriradiatus* in squabs. Jour. Am. Vet. Med. Assn. 79:393.

Le Roux, P. L.: 1926. Helminths collected from the domestic fowl (*Gallus domesticus*) and the domestic pigeon (*Columba livia*) in Natal. 11th–12th Rep. Director Vet. Educ. and Res., Dept. Agr. Union South Africa, Pretoria, pt. I, Sept.:209.

———: 1930. Helminthiasis of domestic stock in the Union of South Africa. Jour. South African Vet. Med. Assn. 1:43 (Oct.).

Levine, P. P.: 1936. The treatment of ascariasis in chickens. Cornell Vet. 26:120.

———: 1938. Infection of the chicken with *Capillaria columbae* (Rud.). Jour. Parasit. 24:45.

Madsen, H.: 1945. The species of *Capillaria* parasitic in the digestive tract of Danish gallinaceous and anatine game birds. Danish Rev. Game Biol. 1:1.

McCulloch, E. C., and Nicholson, L. G.: 1940. Phenothiazine for the removal of *Heterakis gallinae* from chickens. Vet. Med. 35:398.

Olivier, L. J.: 1943. The occurrence of *Syngamus trachea* in mature chickens. Proc. Helminth. Soc. Wash. 10:87.

Ortlepp, R. J.: 1923. The life-history of *Syngamus trachealis* (Montagu) v. Siebold, the gapeworm of chickens. Jour. Helminth. 1:119.

Price, E. W.: 1929. Acanthocephalid larvae from the esophagus of turkey poults. Jour. Parasit. 15:290.

Ransom, B. H.: 1921. The turkey an important factor in the spread of gapeworms. U.S.D.A:, Bul. 939:1.

Riley, W. A., and James, L.: 1922. Life history and methods of control of the chicken nematode (*Heterakis papillosa*, Bloch). 30th Ann. Rep., Minn. Agr. Exper. Sta. (1921–22) :70.

Roberts, F. H. S.: 1937. Studies on the life history and economic importance of *Heterakis gallinae* (Gmelin, 1790; Freeborn, 1923), the caecum worm of fowls. Australian Jour. Exper. Biol. and Med. Sci. 15:429.

Sanders, D. A.: 1929. Manson's eyeworm of poultry. Fla. Agr. Exper. Sta., Bul. 206:567.

Schlegel, M.: 1921. Echinorhynchus polymorphus Brems., seuchenhaftes Entensterben verursacherd. Arch. wiss. u. prakt. Tierheilk. 47:216.

Shillinger, J. E.: 1942. Diseases of farm-raised game birds. U.S.D.A. Yearbook:1230.

Stevenson, E. C.: 1904. A new parasite (*Strongylus quadriradiatus* n. sp.) found in the pigeon. Bur. An. Ind., U.S.D.A., Cir. 47:1.

Sugimoto, M., and Nishiyama, S.: 1937. On the nematode, *Tropisurus fissispinus* (Diesing, 1861), and its transmission to chickens in Formosa. Jour. Jap. Soc. Vet. Sci. 16:305.

Swales, W. E.: 1933. *Tetrameres crami* sp. nov., a nematode parasitizing the proventriculus of a domestic duck in Canada. Canad. Jour. Res. 8:334.

Stoddard, H. L.: 1931. The Bobwhite Quail; Its Habits, Preservation, and Increase. Charles Scribner's Sons, New York, 559 pp.

Taylor, E. L.: 1938. An extension to the known longevity of gapeworm infection in earthworms and snails. Vet. Jour. 94:327.

Tyzzer, E. E.: 1926. *Heterakis vesicularis* Frölich, 1791; a vector of an infectious disease. Proc. Soc. Exper. Biol. and Med. 23:708.

————: 1928. Entero-hepatitis in turkeys and its transmission through the agency of *Heterakis vesicularis*. Rep. Proc. 3rd World's Poultry Cong., p. 286.

Uribe, C: 1922. Observations on the development of *Heterakis papillosa* Bloch in the chicken. Jour. Parasit. 8:167.

Van Valkenburg, H. L.: 1938. Check list of parasites found among principal domestic animals in Puerto Rico. Proc. Helminth. Soc. Wash. 5:7.

Venard, C.: 1933. Helminths and coccidia from Ohio bobwhite. Jour. Parasit. 19:205.

Vigueras, P.: 1929. Una enfermedad parasitaria epizootica de las palomas. Agr. y. zootec. 8:167.

————: 1931. Nota sobre algunos helmintos de *Meleagris gallo pavo*, encontrados en Cuba, con descripcion de una nueva especie. Habana, Cuba, (2) pp.

Walker, H. D.: 1886. The gapeworm of fowls (*Syngamus trachealis*): The earthworm (*Lumbricus terrestris*), its original host. Also, on the prevention of the disease in fowls called the gapes, which is caused by this parasite. Bul. Buffalo Soc. of Nat. Sci. 5:17.

Ward, J. W.: 1945. A new locality record for five species of helminth parasites of the bobwhite quail. Proc. Helminth. Soc. Wash. 12:71.

Wehr, E. E.: 1936. Earthworms as transmitters of *Capillaria annulata*, the "cropworm" of chickens. No. Am. Vet. 17:18.

————: 1937a. Relative abundance of crop worms in turkeys. Macroscopic differentiation of species. Vet. Med. 32:230.

————: 1937b. Observations on the development of the poultry gapeworm, *Syngamus trachea*. Trans. Am. Micr. Soc. 56:72.

————: 1939a. Studies on the development of the pigeon capillarid, *Capillaria columbae*. U.S.D.A., Tech. Bul. 679:19.

————: 1939b. Domestic fowls as hosts of the poultry gapeworm. Poultry Sci. 18:432.

————, Harwood, P. D., and Schaffer, J. M.: 1939. Barium antimonyl tartrate as a remedy for the removal of gapeworms from chickens. Poultry Sci. 18:63.

Wickware, A. B.: 1922. Notes on the parasites of domesticated fowls in Canada. Canad. Vet. Record 3:142.

Wilcox, E. V., and McClelland, C. K.: 1913. Eyeworm of chickens. Hawaii Agr. Exper. Sta., Bul. 43:1.

Yeatter, R. E.: 1934. The Hungarian partridge in the Great Lakes region. Univ. Michigan, School Forestry and Conserv., Bul. 5:92.

# CHAPTER THIRTY-THREE

## CESTODES OF POULTRY

By EVERETT E. WEHR, *Zoological Division, Bureau of Animal Industry,*
*United States Department of Agriculture, Washington, D. C.*

☆   ☆   ☆

The cestodes or tapeworms are flattened, ribbon-shaped, usually segmented worms. As adults, they are found principally in the intestines of their hosts. These worms are hermaphroditic and lack both a mouth and an alimentary canal.

The class **CESTODA** has customarily been divided into three orders, the CESTODARIA, PSEUDOPHYLLIDEA and CYCLOPHYLLIDEA. The CESTODARIA are parasites chiefly of the intestine or body cavity of fishes. They have simple, unsegmented bodies containing a single set of reproductive organs. The scolex is indefinite or absent. These worms resemble the trematodes in appearance, but lack the alimentary tract, and differ from the true tapeworms in having only a single set of reproductive organs. The PSEUDOPHYLLIDEA are, as adults, parasitic in mammals, birds, and reptiles, and in the larval stages are found in copepods and fishes. This order consists of a number of families, of which only one, the Diphyllobothriidae, contains species of economic and medical importance. The CYCLOPHYLLIDEA, or taenioid cestodes are, as adults, parasitic chiefly in the higher vertebrates and are of considerable economic and medical importance. These tapeworms are characterized by having a scolex with four cupshaped suckers and with or without a rostellum.

The taenioid cestodes are grouped into a number of families, four of which contain species infesting poultry. The worms of the family Anoplocephalidae possess neither rostellum nor hooks. The proglottids are usually wider than long, and each proglottid contains one or two sets of genital organs. The genital pores are marginal, and the eggs frequently contain "pyriform" bodies. The species *Aporina delafondi* belongs to this family. The family Davaineidae is composed of tapeworms having a scolex with a simple rostellum which is armed with one or more rows of numerous hammershaped hooks. The suckers are usually also provided with hooks. Each proglottid contains one or two sets of genital organs. The uterus is persistent and saclike, or replaced by either numerous egg capsules or a paruterine body which later becomes transformed into a single egg capsule. Poultry tapeworms of the genera Davainea and Raillietina belong to this family. In the family Dilepididae, the rostellum is usually armed, but the suckers are un-

armed. The genital pores are marginal, one or two in each segment. The uterus is saclike, or resolved into egg capsules—uterus with or without paruterine body. The poultry tapeworms, *Amoebotaenia sphenoides* and *Choanotaenia infundibulum,* belong to this family. The Hymenolepididae is characterized by having a scolex with rostellum usually armed with a single row of hooks; the suckers are unarmed. The genital pores are usually unilateral, rarely two in each segment. The uterus is usually persistent and saclike. The eggs are enclosed in three envelopes. The species of poultry tapeworms belonging to the genus Hymenolepis belong to this family.

**General morphology.** Structurally, a complete tapeworm consists of a head, neck, and a variable number of segments or divisions. The head or scolex of a taenioid tapeworm consists of four cuplike organs or suckers which surround a terminal retractile organ known as the rostellum. Hooks may or may not be found on the rostellum, and deciduous spines often arm the suckers. The number, size, and shape of the rostellar hooks vary as to species, and these variations are used by systematists in differentiating species and even genera of tapeworms. The term, neck, is applied to the narrowed and unsegmented region located just back of the head. Since this region in some cestodes is not macroscopically distinct from the head, the neck region has been considered to be absent in those species. The segments or divisions of a tapeworm when taken collectively are generally spoken of as the strobila, and each segment or division as a proglottid. The size, shape, and development of proglottids vary tremendously even in the same individual worm. The anterior segments are usually broader than long and contain few, if any, recognizable internal organs. Those segments near the middle of the body of the tapeworm may have the antero-posterior diameter proportionately greater than that of the anterior segments. These segments are spoken of as mature segments, since in these proglottids both the male and female reproductive organs are distinctly differentiated. Eggs are not usually found in segments of this part of the body. The terminal or gravid segments are variable in shape and usually contain the uteri and eggs or only eggs with the uterus either partly or wholly obliterated.

Since tapeworms lack an alimentary canal, food is absorbed through the surface of the body.

A tapeworm grows from the neck backwards, and segments are continually being budded off from the proliferating tissue found in this region. Therefore, the segments farthest removed from the growing region are the oldest from the standpoint of development. The newly-formed segment contains no distinguishable organs, while the terminal segments of a completely formed tapeworm may be nothing more than egg sacs. The latter are known as gravid segments and are the ones usually found in the droppings of infested birds.

All adult tapeworms of poultry are found usually in the small intestines

of their hosts. However, *Hymenolepis megalops,* the large-headed tapeworm of ducks, occurs in the cloaca and bursa Fabricii of these birds. Each species of tapeworm usually shows some predilection for a particular portion of the small intestine to which to attach. The species *Hymenolepis carioca, H. cantaniana, Amoebotaenia sphenoides,* and *Davainea proglottina* are usually found in the duodenal region of the small intestine; *Raillietina cesticillus, Choanotaenia infundibulum,* and *Metroliasthes lucida* in the jejunal region; and *Raillietina tetragona* and *R. echinobothrida* in the distal portion or ileum. However, in heavy infestations, tapeworms may be found in portions of the small intestine other than their more normal locations.

Adult tapeworms of poultry differ considerably as to length and as to number of proglottids or segments. *Davainea proglottina* and *Amoebotaenia sphenoides* are two of the smallest tapeworms found in poultry. Mature specimens of the former species measure up to 4 mm. in length and have a range of segments from four to nine, while those of the latter species reach a length of from 2 to 3.5 mm. and possess approximately thirty proglottids. *Raillietina echinobothrida* and *R. tetragona,* on the other hand, are two of the largest tapeworms infesting poultry. Mature specimens of both of these tapeworms may attain a length of approximately 25 cm. and possess large numbers of proglottids.

**Development.** In the case of every tapeworm of poultry in which the life history is known, an intermediate host is necessary for the completion of the life cycle. Investigations have shown invariably that intermediate hosts of tapeworms have always been found to be invertebrates, such as a beetle, fly, snail, slug, or crustacean. Tapeworm segments are devoured by dung-feeding insects either along with their normal food or because they are attracted to the attention of the invertebrates by their movements.

The type of intermediate host that serves a particular tapeworm in its successful transference from one bird host to another depends to a large degree on the habits of the avian host. In case of terrestrial birds, such as chickens, turkeys, guinea fowls, etc., which deposit their body wastes principally away from ponds and streams, the intermediate hosts must necessarily have to be forms of animal life that lead a terrestrial life, or at least an amphibious one. On the other hand, tapeworms inhabiting water birds, such as ducks and geese, usually have aquatic invertebrates as intermediate hosts.

Invertebrates, which serve as intermediate hosts of poultry tapeworms, become infested with larval tapeworms by ingesting along with their food the free eggs or the egg-bearing segments voided by the infested birds. Following ingestion the eggs hatch in the digestive tract, the embryos or larvae penetrate the intestinal wall, enter the body cavity, and after a few days become transformed into small, white, bladder-like, spherical bodies, known as cysticercoids (small cysts) . These cysts are distinctly visible to the unaided eye when placed in water after removal from the body of the intermediate

host.  Under proper magnification the head of the tapeworm may be seen near the center of the cyst.

Approximately three weeks are required for the embryos to develop into the cysticercoid stage after the eggs have been ingested by the intermediate host. No further development of the tapeworm takes place in the invertebrate host. The cysticercoids may remain alive in the invertebrate host and infective to the bird host for many months.

Poultry become infected with tapeworms by swallowing, with their food and water, insects, snails, slugs, and other forms of animal life that may serve as intermediate hosts of these parasites.  The cysticercoid is freed from the body of the intermediate host by the action of the digestive juices.  Soon after the cysticercoid is liberated the head evaginates and becomes attached to the intestinal wall.  New segments or proglottids begin to form immediately at the neck region, and within approximately three weeks a mature tapeworm is formed.  The entire life cycle, therefore, takes about six weeks for completion, but under unfavorable conditions a longer period of time may be necessary.

**Gross pathology.**  A few tapeworms may produce little or no perceptible gross pathological changes in the intestines.  However, in heavy tapeworm infestations, a more or less extensive catarrhal enteritis and diarrhea may result.  At least one species of tapeworm, *Raillietina echinobothrida,* causes the formation of nodules in the intestinal wall.  Inasmuch as this condition closely resembles tuberculosis, it is important that the two conditions be kept in mind in attempting to arrive at a diagnosis of the condition present.  The presence of intestinal nodules—sometimes distinctly visible on the outer surface of the intestinal wall—and the absence of tapeworms is strongly suggestive of tuberculosis.  However, a diagnosis of intestinal tuberculosis should not be made without first eliminating tapeworms of this species as a cause of the nodules.  Mature specimens of this tapeworm are usually several centimeters long, but observations have shown that, in many cases, infestations involved individual tapeworms containing only a very few segments, sometimes only the heads.  In such cases, the parasites may be easily overlooked if only a casual or hurried examination is made.  In doubtful cases, the intestine should be scraped with a scalpel or other suitable instrument and a careful examination made of the scrapings under suitable magnification for the presence of small tapeworms or their heads.  The presence of tapeworms and the absence of tubercles in the liver and other organs is indicative of tapeworm disease.  Another species of tapeworm, *Raillietina tetragona,* which is morphologically very similar to *R. echinobothrida* and often confused with it, has not been definitely associated with tuberculosis-like lesions.

Leg weakness and paralysis have frequently been attributable to tapeworm infestation.  However, the relationship of tapeworms to these diseases

is still unknown. Should these conditions be intimately associated with the presence of tapeworms, the mere removal of the parasite should clear up the condition. The fact that birds which had previously shown symptoms of leg weakness and paralysis were free from tapeworms at necropsy seems to disprove the idea that tapeworms are in a large degree responsible for these conditions. Capillary congestion; lymphocyte, polymorphonuclear, and eosinophil infiltration; proliferation of epithelium; and fibrosis are other conditions which have been associated with tapeworm infestations.

There is some evidence to show that birds heavily parasitized with tapeworms are not as productive as uninfested ones. Under ordinary conditions birds may tolerate a fairly heavy tapeworm infestation, at least for a time. However, young birds and hens in heavy production do not fare so well when heavily infested with these parasites.

**Importance of cestodes as parasites of poultry.** Chickens in this country may be infested with one to as many as seven species of tapeworms. With the exception of one or two of these species, all are of common occurrence. A few years ago, the list of intermediate hosts of poultry tapeworms was small, but investigations within the last few years have been responsible for an alarming increase in the number of invertebrate intermediate hosts that tapeworms of poultry may utilize for the development of their larvae.

The control of poultry tapeworms involves treatment for the removal of the tapeworms themselves and the reduction of the numbers of intermediate hosts by sanitary measures. Since the treatment of fowl taeniasis is still in an unsatisfactory state, sanitation has almost wholly been relied upon to prevent tapeworm infestation. This method of control involves first of all the proper disposal of poultry manure containing the eggs of tapeworms so that the intermediate hosts cannot become infected with the larval stages of these parasites. Many of the intermediate hosts are flying insects, and once the latter have become infected with larval tapeworms they may be responsible for the spread of the disease to distant flocks. Recent investigations have indicated that clean birds held in close proximity to infested birds will invariably become infected with tapeworms within a relatively short time.

The species of cestodes parasitizing poultry of the United States belong to four families which may be differentiated by the following key:

1. Head armed with numerous hammer-shaped hooks  .   .      Davaineidae
   Head armed with hooks not hammer-shaped, or unarmed   .   .   .   2
2. Testes few, 1 to 4, rarely more .   .   .   .   .   .      Hymenolepididae
   Testes numerous, more than 4 .   .   .   .   .   .   .   .   .   3
3. Head lacking rostellum; no paruterine organs in species occurring in poultry   .   .   .   .   .   .   .   .   .   .   .      Anoplocephalidae
   Head with retractile rostellum, usually armed, or, rarely, unarmed; rarely without rostellum; with or without paruterine organs   .   Dilepididae

## LIST OF TAPEWORMS KNOWN FROM POULTRY OF UNITED STATES

The following is a list of the species of tapeworms found in poultry of this country, with their primary and secondary hosts, usual location and kinds of poultry affected.

| Tapeworms | Location | Intermediate hosts | Definitive hosts |
|---|---|---|---|
| Davainea proglottina | Duodenum | Slugs, snails | Chicken |
| Davainea meleagridis | Duodenum | Unknown | Turkey |
| Amoebotaenia sphenoides | Duodenum | Earthworms | Chicken, Turkey |
| Hymenolepis carioca | Duodenum | Stable fly Dung beetles | Chicken, Turkey Bobwhite quail |
| Hymenolepis cantaniana | Duodenum | Beetles | Chicken, Turkey, Peafowl Bobwhite quail |
| Raillietina cesticillus | Jejunum | Housefly Beetles | Chicken, Turkey Guinea fowl Bobwhite quail Gray jungle fowl |
| Choanotaenia infundibulum | Jejunum | Housefly, Beetles | Chicken, Turkey |
| Raillietina tetragona | Ileum | Ants | Chicken, Turkey Guinea fowl, Peafowl Bobwhite quail |
| Raillietina echinobothrida | Ileum | Ants | Chicken, Turkey |
| Metroliasthes lucida | Ileum | Grasshoppers | Turkey, Chicken Guinea fowl |
| Hymenolepis compressa | Intestine | Unknown | Duck, Goose |
| Hymenolepis introversa | Intestine | Unknown | Duck |
| Hymenolepis megalops | Cloaca and bursa of Fabricius | Unknown | Duck |
| Hymenolepis tritesticulata | Intestine | Unknown | Duck |
| Hymenolepis coronula | Small intestine | Crustaceans Snails | Duck, Goose |
| Hymenolepis lanceolata | Small intestine | Crustaceans | Duck, Goose |
| Hymenolepis tenuirostris | Small intestine | Crustaceans Crayfish | Duck, Goose |
| Raillietina magninumida | Small intestine | Beetles | Guinea fowl |
| Raillietina ransomi | Small intestine | Unknown | Wild turkey |
| Raillietina williamsi | Small intestine | Unknown | Wild turkey |
| Aporina delafondi | Small intestine | Unknown | Pigeon |

## CLASSIFICATION OF POULTRY TAPEWORMS

The tapeworms of poultry belong to the general group designated as taenioid cestodes which are characterized primarily by the presence of four cupshaped suckers upon the head. The following key will aid in the differentiation of the genera of tapeworms found in poultry of this country:

1. Rostellum absent . . . . . . . . . . . . . . 2
   Rostellum present. . . . . . . . . . . . . 3
2. Paruterine organ present . . . . . . . . *Metroliasthes*
   Paruterine organ absent . . . . . . . . . *Aporina*
3. Mature worms small . . . . . . . . . . . . 4
   Mature worms large . . . . . . . . . . . . 5
4. Strobila consisting of 2 to 9 segments . . . . . . *Davainea*
   Strobila consisting of numerous segments . . . . *Amoebotaenia*
5. Testes 3 in number . . . . . . . . . . *Hymenolepis*
   Testes more than 3 in number . . . . . . . . . . 6
6. Rostellum armed with a single row of 16 to 20 hooks, each 20 to 30µ
   long . . . . . . . . . . . . . . *Choanotaenia*
   Rostellum armed with either a single row or double row of 100 or more
   hooks, each 6 to 15µ long . . . . . . . . . *Raillietina*

## DESCRIPTIONS OF POULTRY TAPEWORMS

To facilitate somewhat the identification of the species of poultry tapeworms, they have been grouped according to their normal location within the intestine of the hosts, i.e., duodenum, jejunum, and ileum, with a brief description of each species.

Five species of tapeworms normally inhabit the duodenal region. Three of these species, *Davainea proglottina, D. meleagridis,* and *Amoebotaenia sphenoides* are very small worms, rarely exceeding 5 mm. in length, and possessing relatively few segments. The other species, *Hymenolepis carioca* and *H. cantaniana,* are relatively long worms and are composed of many segments.

### DILEPIDIDAE

Members of this family are characterized by having a single set of reproductive organs in each proglottid. The uterus is saclike and more or less lobed or reticulate. Paruterine bodies are present or absent.

#### *Amoebotaenia sphenoides* (Railliet, 1892)

**Synonyms.** *Taenia cuneata* von Linstow, 1872, not Batsch, 1786; *Taenia sphenoides* Railliet, 1892; *Dicranotaenia sphenoides* (Railliet, 1892) Railliet, 1896.

**Description.** Mature worms 2 to 3.5 mm. long, triangular or roughly fusiform in shape (Fig. 33.1 A). Suckers unarmed; rostellum armed with a single row of 12 to 14 hooks, 25 to 32µ long (Fig. 33.1 B). Genital pores

usually regularly alternate, located at extreme anterior point of segment margin. Testes 12 to 15 in number, usually in a transverse row across posterior part of segment. Eggs (Fig. 33.1 C) not contained in capsules.

This tapeworm, which usually occurs in the duodenal region of the small intestine, is apparently not a common parasite of poultry in the United States. It has been reported from chickens in Kansas by Ferry (1934), from chickens in Texas by Adams and Geiser (1933), and from chickens and turkeys in Michigan by Stafseth (1940).

**Life history.** The intermediate host of this tapeworm is the earthworm. Mönnig (1927) grew the cysticercoids in earthworms (*Ocnerodrilus* (*Ilyogenia*) *africanus*) in 14 days. Four weeks were required for the cysticercoids to develop into adult tapeworms in chickens. Cysticercoids from earthworms were identified as this species by Grassi and Rovelli (1889) and Meggitt (1916). Chickens become infected by eating earthworms which carry the infective larva or cysticercoids of this cestode parasite.

**Pathology.** The damage done by this tapeworm is comparatively slight, according to Meggitt (1926). However, deaths in poultry as being due to this parasite have been reported.



FIG. 33.1. *Amoebotaenia sphenoides.* A—entire worm. (From Mönnig, 1926.) B-rostellar hooks. C—egg, original.

*Choanotaenia infundibulum* (Bloch, 1779)
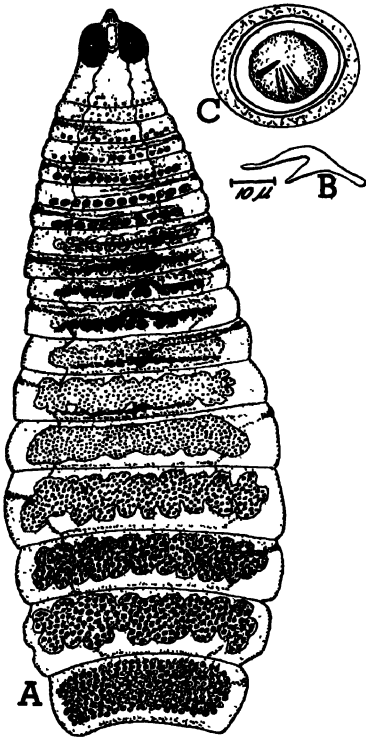
**Synonyms.** *Taenia infundibulum* Bloch, 1779; *Drepanidotaenia infundibuliformis* (Goeze, 1782) Railliet, 1893; *Choanotaenia infundibuliformis* (Goeze, 1782) Railliet, 1896.

**Description.** Mature worms attain a length of 23 cm. Suckers unarmed (Fig. 33.2 A); rostellum armed with a single row of 16 to 20 hooks, occasionally 22, 20 to 30μ long (Fig. 33.2 B). Genital pores irregularly alternate. Testes 25 to 40, occasionally as many as 55 to 60, grouped in posterior part of segment (Fig. 33.2 C). Eggs not contained in capsules (Fig. 33.2 D).

This species may be readily distinguished from the other poultry tapeworms by the rostellum which is armed with a single row of relatively few and very large hooks.

. This cestode inhabits principally the jejunal region of the small intestine

of chickens and turkeys and is widely distributed among these birds in the
United States.

**Life history.** Birds become infected with adults of *C. infundibulum* by
eating house flies, grasshoppers, and several species of beetles. Cysticercoids
have been found in house flies and in some species of beetles as natural infes-
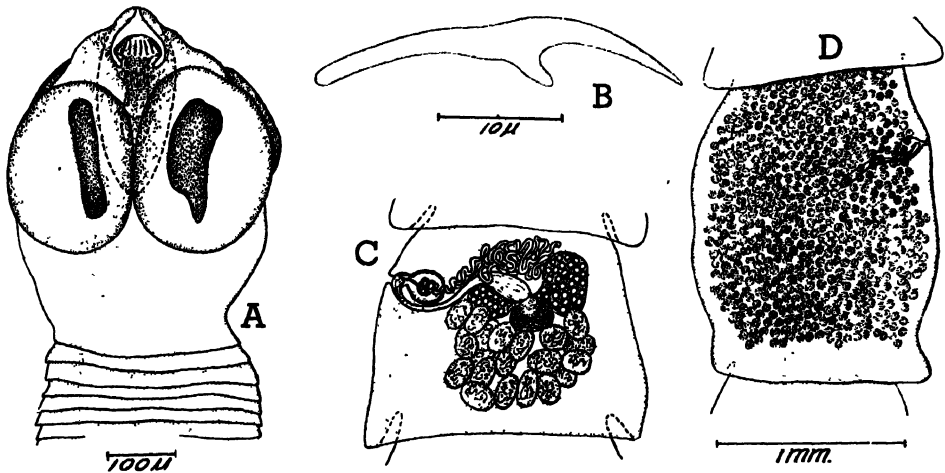


Fig. 33.2. *Choanotaenia infundibulum*. A—scolex. B—rostellar hook. C—mature seg-
ment. D—gravid segment. (From Ransom, 1905.)

tations, and also after the insects have been fed eggs of this tapeworm.
Horsfall and Jones (1937) reported that at a temperature of 75 to 90° F.,
17 to 20 days is the minimum time for development of the cysticercoids to
the infective stage in the grasshopper, *Melanoplus femurrubrum.* At a
temperature of 60 to 75° F., 48 days is the minimum time for the develop-
ment of the cysticercoids in the beetle, *Aphodius granarius.* The adult worm
in the chicken requires from two to three weeks for development to maturity.

**Pathology.** Probably similar to *R. cesticillus.*

### *Metroliasthes lucida* (Ransom, 1900)

**Description.** Mature worms about 20 cm. long. Suckers unarmed, rostel-
lum lacking (Fig. 33.3 A). Genital pores irregularly alternate, near middle
of, or in gravid segments, definitely posterior to middle of segment margin.
Uterus, when fully developed, consisting of two sacs, lying side by side and
very close together in posterior part of segment (Fig. 33.3 D). Paruterine
organ, a conical structure, developing anterior to uterus, eventually becom-
ing a heavy-walled egg capsule for the retention of the eggs (Fig. 33.3 E).

This species is a very common tapeworm of turkeys in this country. It
was reported from a chicken by Ransom (1905), but he evidently doubted
the validity of the host record since he stated that the occurrence of *Metro-*

*liasthes lucida* in chickens is doubtful. However, the occurrence of this species in chickens has been reported more recently by Rietz (1930) from West Virginia, by Southwell (1921) from India, and by Schwartz (1925) from South Africa. It is readily recognized by the large unarmed head and the prominent spherical egg capsule which is easily seen in the posterior part of each of the transparent segments in the posterior part of the body.

**Life history.** Cysticercoids were obtained by Jones (1930) from grass-
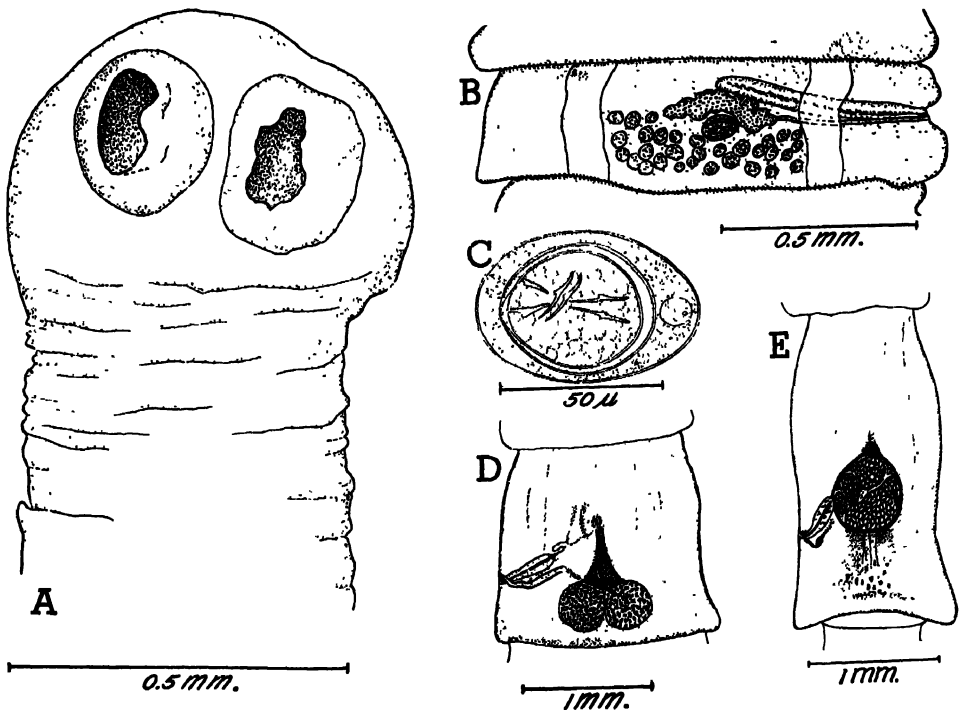


FIG. 33.3. *Metroliasthes lucida.* A—scolex, original. B—mature segment. C—egg. D—segment showing two-part uterus and developing paruterine organ. E—gravid segment. (From Ransom, 1900.)

hoppers several weeks after feeding to the insects gravid segments of *M. lucida;* both laboratory-bred grasshoppers and those collected in the field become infected. Jones (1936) infected turkeys and guinea fowls with *M. lucida* after being fed cysticercoids from grasshoppers (*Melanoplus* species, *Chorthippus curtipennis,* and *Paroxya clavuliger*) ; chicks and quail remained negative for tapeworms after being fed cysticercoids of *M. lucida* from grasshoppers or beetles. The time required for the development of the cysticercoids in the insect host varies from two to six weeks. Approximately three weeks are required for the development of the adult worm to maturity in the avian host.

**Pathology.** Probably similar to that of *R. cesticillus.*

Tapeworms of this family have a scolex with a simple rostellum which is armed with one or more rows of numerous hammer-shaped hooks. The suckers may be armed or unarmed. One or two sets of reproductive organs may be present in each segment. The uterus is persistent and saclike, or replaced either by numerous egg capsules or by a paruterine body transforming later into a single egg capsule.

*Davainea proglottina* (Davaine, 1860)

**Synonym.** *Taenia proglottina* Davaine, 1860.

**Description.** Mature worms attain a length of about 4 mm. (Fig. 33.4 A). The strobila consists of from 2 to 5 segments, rarely as many as 9. Each succeeding segment gradually increases in length and breadth, the last segment often being larger than the remainder of the parasite. Suckers armed with 3 to 6 rows of small hooklets, 5 to 8µ long. Genital pores usually regularly alternate, located at extreme anterior point of segment margin. Testes 12 to 21 in number (Fig. 33.4 B). One egg in each egg capsule.

In the United States, this tapeworm is not as cosmopolitan in its distribution as some of the other cestodes of poultry, being found chiefly in the moister parts. It has been reported from both the eastern and western coastal states.

**Life history.** Cysticercoids of this tapeworm develop in approximately three weeks in snails and slugs. Levine (1938) experimentally infected the garden slug (*Agriolimax agrestis*) with cysticercoids of *D. proglottina* and, in turn, infected chickens by feeding them garden slugs containing mature cysticercoids. When infested slugs or snails are eaten by chickens, the infective larva or cysticercoid develops to the adult worm with 4 segments in approximately 8 days.



Fig. 33.4. *Davainea proglottina.* A—entire worm, with eggs in last segment. B — mature segment. (From Meggitt, 1926.)

**Pathology.** This tapeworm has been considered to be one of the obviously dangerous tapeworms of poultry. It has been observed that infested birds become emaciated and dull, the plumage becomes dry and ruffled, the movements slow, and the breathing rapid. At necropsy, the intestinal mucosa

appears thickened, which may be hemorrhagic, and the intestine may contain a large quantity of mucus, which tends to be fetid. Crawley (1922) has reported this worm as killing chickens in Pennsylvania. Rietz (1930) has reported paralysis associated with the presence of this worm. However, the true relationship of leg weakness to this disease is still unknown.

### *Davainea meleagridis* (Jones, 1936)

**Description.** Mature specimens up to 5 mm. long, composed of 17 to 22 segments. Suckers armed with 4 to 6 rows of hooklets, the longest about 5μ long; rostellum with a double row of about 100 to 130 hooks, 8 to 10μ long. Genital pores usually regularly alternate, located in extreme anterior point of segment margin. Testes 20 to 26 in number, in posterior half of segment. One egg in each capsule.

This parasite was described from the duodenum of the domestic turkey by Jones (1936) in the vicinity of Washington, D. C.

**Life history.** Unknown.

**Pathology.** Unknown.

### *Raillietina cesticillus* (Molin, 1858)

**Synonyms.** *Taenia cesticillus* Molin, 1858; *Raillietina cesticillus* (Molin, 1858) Joyeux, 1923.

**Description.** Mature worms may attain a length of as much as 12 cm. Suckers unarmed; rostellum armed with two rows of hooks, about 300 to 500 in number (Fig. 33.5 A and B). Genital pores irregularly alternate, located anterior to middle of segment margin. Testes 16 to 30 in number, in posterior part of segment (Fig. 33.5 C). Uterus divided into egg capsules, each capsule containing a single egg.

The most distinctive feature of this tapeworm is the unusually broad and flattened rostellum, with 2 rows of hooks near its base.

This fowl cestode is probably one of the most common species occurring in poultry. It is a rather large species, and its habitat is the duodenal and jejunal regions. Southwell (1930) reported *R. cesticillus* from *Gallus sonnerati*, the gray jungle fowl, in the Zoological Gardens of Calcutta.

**Life history.** Birds become infected with *R. cesticillus* after being fed various infested ground beetles and dung beetles. Cysticercoids have been observed in such beetles as *Anisotarsus* spp., *Amara* spp., *Anaferonia* spp., *Harpalus* spp., *Pterostichus* spp., and other ground and dung beetles after they have been given experimental feedings of gravid segments of *R. cesticillus*, and also have been observed in natural infestations in some of these beetles. Larva in beetles apparently requires from three to four weeks to develop to a stage infective for chickens. Adult worms in primary host usually require from two to three weeks to develop to maturity.

**Pathology.** This worm has been reported to cause degenerations and

inflammations of the villi of the intestine at the point of attachment by the rostellum. Heavy infestations in young birds may cause emaciation. However, Stoddard (1931) noted no serious inflammation of the intestinal walls, nor was stoppage of the intestines found to result from the presence of the worms in quail. Ackert and Reid (1937) and Ackert (1932) demonstrated experimentally that chickens two and one-half to five months of age are more resistant to infestation with this species of tapeworm than younger birds, and that a reduction in the blood sugar and hemoglobin contents of the blood
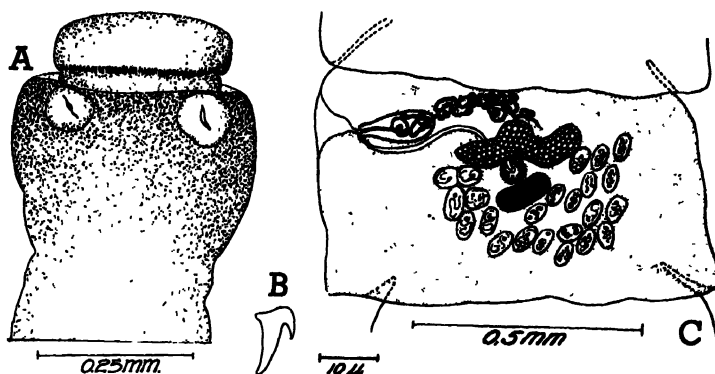


Fig. 33.5. *Raillietina cesticillus.* A—head, original. B—hook from rostellum. C—mature segment. (From Ransom, 1905.)

resulted from such infestations. Harwood and Luttermoser (1938) reported that the growth rates of Rhode Island Red and White Leghorn chicks were retarded by infestations with *R. cesticillus.*

### *Raillietina echinobothrida* (Megnin, 1881)

**Synonyms.** *Taenia echinobothrida* Megnin, 1881; *Raillietina echinobothrida* (Megnin, 1881) Fuhrman, 1924.

**Description.** Mature specimens measure up to 25 cm. long. Suckers armed with 8 to 15 rows of hooks, 5 to 15µ long; rostellum armed with 2 rows of 200 to 240 hooks, 10 to 14µ long (Fig. 33.6 A and C). Genital pores almost unilateral, or definitely irregularly alternate, located at middle or, usually, posterior to middle of segment margin (Fig. 33.6 B). Testes 20 to 30, occasionally as many as 45 in number. Uterus ultimately forming egg capsules, each capsule usually containing a single egg. Posterior segments of strobila frequently becoming constricted longitudinally through median line to form windows in the center of the segments. However, this appearance of the gravid segments is not constant in all specimens.

*Raillietina echinobothrida* is apparently widely distributed among poultry.

**Life history.** Jones and Horsfall (1935) reported that the ants, *Tetramorium caespitum* and *Pheidole vinelandica,* naturally harbored bladderworms or cysticercoids of *R. echinobothrida* and also those of another closely related species, *Raillietina tetragona.* The cysticercoids of the two species were fed to laboratory-reared chickens. Three weeks after feeding the cysticercoids, adults' of the two species of tapeworms were recovered postmortem from the experimentally fed birds; the controls were negative. All
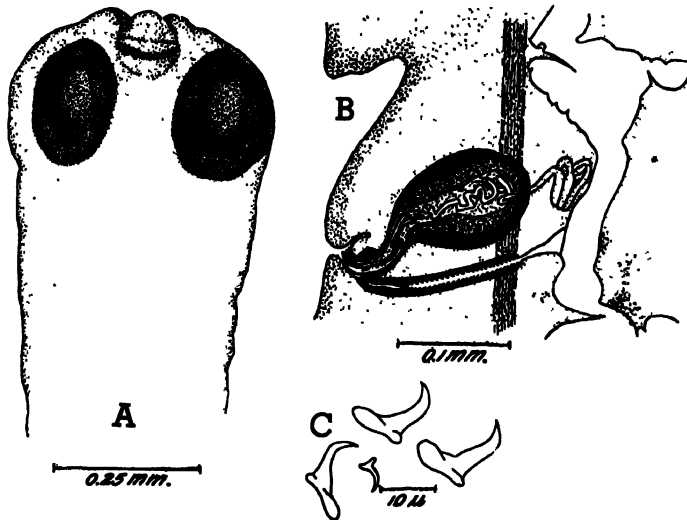


Fig. 33.6. *Raillietina echinobothrida.* A—scolex, original. B—section through region of genital pore showing cirrus pouch and part of vagina. (From Lang, 1929.) C—hooks from suckers.

attempts to produce experimental infections in ants were unsuccessful. Large numbers of undissected ants collected from infected poultry runs were fed to twenty-three chickens; nineteen of the chickens later became infected. Joyeux and Baer (1937) reported finding cysticercoids of *R. echinobothrida* in naturally infested ants, *Tetramorium semileave,* in the region of Marseilles, France.

**Pathology.** This worm causes the formation of tubercles on the intestinal wall of infested birds (Fig. 33.7). This condition resembles tuberculosis and, therefore, must be differentiated from that disease.

Gage and Opperman (1909) reported losses of 50 per cent in affected flocks in Maryland. They noted emaciation and a mucoid diarrhea as early symptoms, and later listlessness, loss of appetite, and a tendency to huddle; some birds are weak and epileptic. Death comes suddenly, accompanied by convulsions.

*Raillietina tetragona* (Molin, 1858)

**Synonyms.** *Taenia tetragona* Molin, 1858; *Raillietina tetragona* (Molin, 1858) Joyeux, 1927.

**Description.** Worms measure as much as 25 cm. long. Suckers armed with 8 to 12 rows of small hooks, 3 to 8µ long; rostellum armed with about 90 to 130 hooks, 6 to 8µ long, arranged in 1 or 2 rows (Fig. 33.8 A). Genital pores usually unilateral, rarely irregularly alternate, located anterior to



Fig. 33.7. Nodular disease of intestine of chicken caused by tapeworms *Raillietina echinobothiida*. (After Bushnell and Brandly, 1929.)

middle of segment margin. Testes 18 to 35 in number (Fig. 33.8 D). Uterus eventually breaking up into egg capsules, 6 to 12 eggs in each capsule (Fig. 33.8 B).

This worm is morphologically very similar to *Raillietina echinobothrida*. It is of common occurrence but is rarely associated with the distinct tuberculosis-like lesions produced by the former species.

**Life history.** See life history of *R. echinobothrida*.

**Pathology.** Lopez-Neyra (1931) reported a single case in which he found small nodules in the intestine due to this species. In quail Stoddard (1931) observed that this species may be the principal or only cause of death in

cases of heavy infestations. Of twenty-five birds, the deaths of which were attributed to infestation with this species, the youngest was 17 days old, and the oldest 60 days; the greatest mortality occurred between the ages of 25 and 40 days. Although many birds may recover if they survive to two months of age, they are almost certain to be under-sized. Quail heavily infested with specimens of this tapeworm almost invariably have their crops and gizzards filled with food. That portion of the intestine occupied by these tapeworms
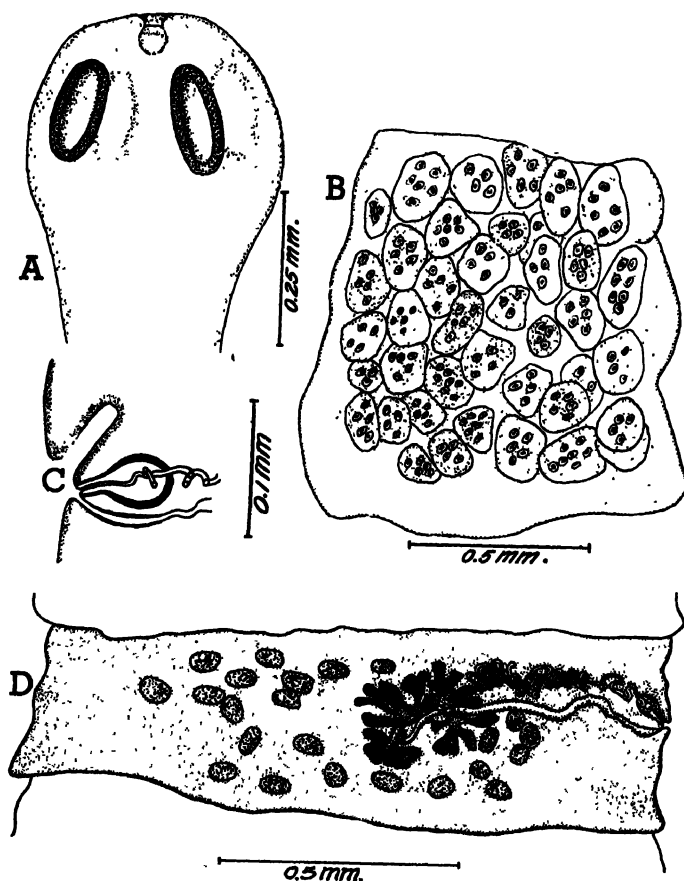


FIG. 33.8. *Raillietina tetragona.* A—scolex. B—gravid segment. Original. C—pore region showing cirrus pouch and vagina. (After Lang, 1929.) D—mature segment. (After Ransom, 1905.)

sometimes becomes so distended that it is reduced to nearly one-half its length, being thrown into ridges of a purplish red color. The lining of the intestine frequently sloughs off in case of heavy infestations. In several instances Stoddard observed that bobwhites heavily infested with this species moved with difficulty, a parital paralysis being evident.

## Raillietina magninumida (Jones, 1930)

**Synonym.** *Raillietina (Paroniella) magninumida* Jones, 1930.

**Description.** Mature worms about 6 to 15 cm. long. Suckers armed with about 10 rows of hooks, the largest 7 to 8μ long; rostellum armed with two rows of about 150 to 170 hooks, 8 to 11μ long (Fig. 33.9 A). Genital pores unilateral. Testes 12 to 20 in number (Fig. 33.9 B). Egg capsules containing one egg each.
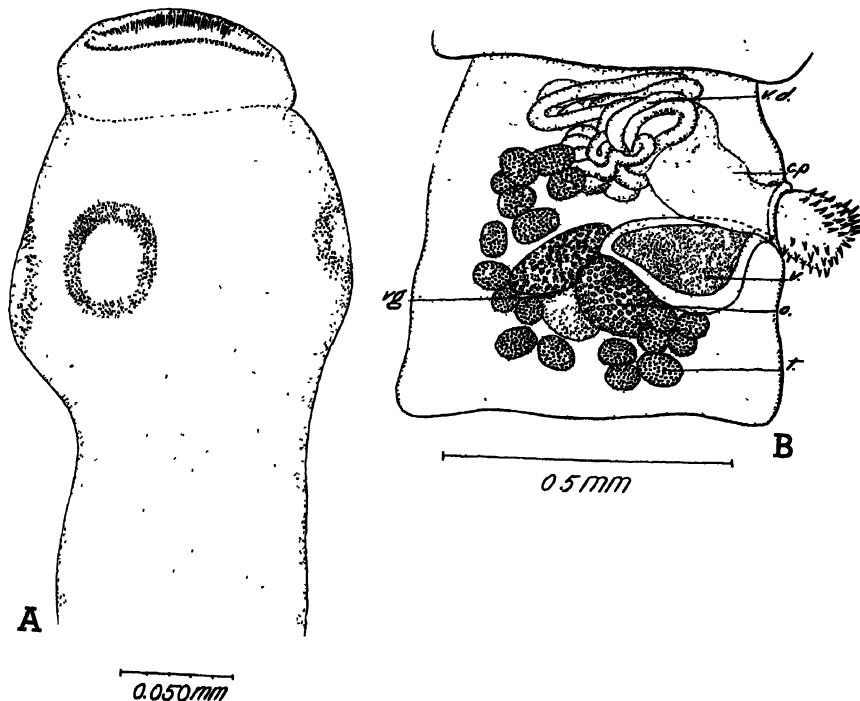


FIG. 33.9. *Raillietina magninumida.* A—scolex with rostellum extended. B—mature segment (c.p., cirrus pouch, o.—ovary, t.—testes, v.—vagina, v.g.—viteline gland, v.d.—vas deferens). Original.

This tapeworm has been reported from guinea fowls in the District of Columbia and Maryland. Hudson (1934) considered *R. magninumida* as a synonym of *R. numida* (Fuhrmann, 1912). The latter species occurs in the guinea fowl of Africa.

**Life history.** Guinea fowls become infected with this species by ingesting beetles carrying cysticercoids of this tapeworm. Cysticercoids have been found in beetles both as a result of the experimental feeding to them of gravid tapeworm segments, and in natural infestations. Approximately three weeks are required for the larva to develop to the infective stage in the beetle, and three weeks more are necessary for the cysticercoid to develop to the adult form in the guinea fowl.

**Pathology.** Adult birds seem little affected by this species, but younger birds appear to be considerably weakened by heavy infestations. Detailed pathology of this species has not been studied.

## Raillietina ransomi (Williams, 1931)

**Synonym.** *Davainea ransomi* Williams, 1931.

**Description.** Mature worms from 4 mm. to 1.4 cm. long by 650μ to 1.14 mm. wide. Total number of segments varying from 24 to 61, usually between
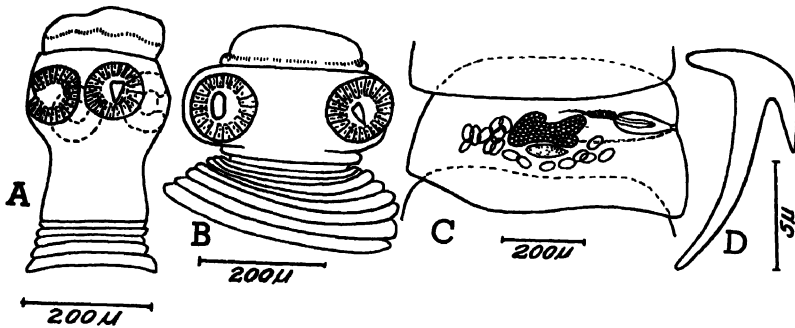


Fig. 33.10. *Raillietina ransomi.* A—head fully extended. B—head partially contracted. C—mature segment. D—hook. (From Williams, 1931.)

30 and 40. Suckers unarmed, round or slightly oval, 85 to 100μ in diameter (Fig. 33.10 A and B). Rostellum well developed, 53 to 91μ long and 150 to 206μ wide, hooks 500 to 520, in 2 rows, 8.8 to 9.6μ long and 11.2 to 12μ long (Fig. 33.10 D). Genital pores irregularly alternate, anterior to middle of segment margin. Testes 15 to 25 in number (Fig. 33.10 C). Uterus at first saclike, then branched, and finally disintegrating, the "embryos" being scattered through the parenchyma.

This species of tapeworm was reported by Williams (1931) and Wehr and Coburn (1943) from the eastern wild turkey.

**Life history.** Unknown.

**Pathology.** Unknown.

## Raillietina williamsi Fuhrmann, 1932

**Synonyms.** *Davainea fuhrmanni* of Williams, 1931, not Southwell, 1922; *Raillietina (Raillietina) williamsi* Fuhrmann, 1932.

**Description.** Mature specimens about 14.3 to 36.7 cm. long by 3.5 to 4.25 mm. wide. Suckers ellipsoidal, 150 to 190μ long by 135 to 170μ wide, armed with instable hooks, very deciduous, in 12 to 13 rows, those of the outer row being largest (Fig. 33.11 A). Rostellum hemispherical, 200 to 214μ in diameter, armed with double crown of 152 to 156 hooks, larger and smaller hooks alternating (Fig. 33.11 B). Genital pores unilateral, in an-

terior thirds of segment margin (Fig. 33.11 C). Uterus breaking up into 75 to 100 egg capsules, each with 8 to 13 eggs (Fig. 33.11 D).

This tapeworm occurs commonly in the wild turkey.

**Life history.** Unknown.

**Pathology.** Unknown.

### ANOPLOCEPHALIDAE

These worms lack both rostellum and hooks. The proglottids are usually
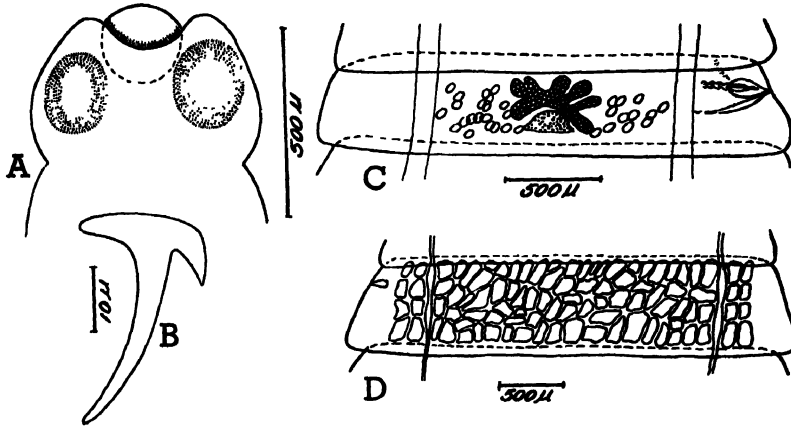


FIG. 33.11. *Raillietina williamsi.* A—head with rostellum partially retracted. B—rostellar hooks. C—mature segment. D—gravid segment showing a single layer of egg capsules. (From Williams, 1931.)

wider than long, and each contains one or two sets of reproductive organs. The testes are numerous. The uterus may persist or be replaced by egg capsules, or the eggs may pass into one or more paruterine organs. Eggs contain "pyriform bodies."

### *Aporina delafondi* (Railliet, 1892)

**Synonyms.** *Taenia delafondi* Railliet, 1892; *Bertiella delafondi* (Railliet, 1892) Railliet and Henry, 1909.

**Description.** Mature worms 7 to 16.5 cm. long. Suckers unarmed; rostellum absent. Genital pores irregularly alternate, located in anterior third of segment margin. Testes about 100 in number. Eggs not contained in capsules.

This is a common tapeworm of pigeons in several parts of the world. In the United States, it has been collected from pigeons in Iowa and Texas.

**Life history.** Unknown.

**Pathology.** Unknown.

### HYMENOLEPIDIDAE

The hymenolepid tapeworms have a scolex with rostellum that is armed

with one row of hooks, rarely with a double row, or unarmed. The number of testes rarely more than four. The uterus is saclike, rarely reticulate. The eggs are enclosed in three envelopes.

## *Hymenolepis carioca* (Magalhães, 1898)

**Synonyms.** *Davainea carioca* Magalhães, 1898; *Weinlandia carioca* Mayhew, 1925.
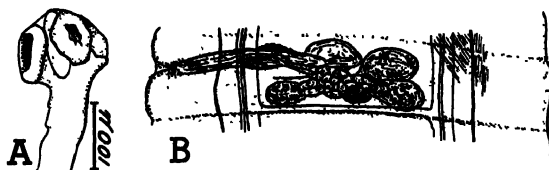


FIG. 33.12. *Hymenolepis carioca.* A—scolex. B—mature segment. (After Ransom, 1902.)

**Description.** Mature specimens 3 to 8 cm. long, composed of many hundreds of segments; segments 3 to 5 times broader than long. Suckers and rostellum unarmed (Fig. 33.12 A). Genital pores unilateral, located anterior to middle of segment margin. Testes 3 in number, usually in a more or less straight row across the segment (Fig. 33.12 B).

This tapeworm is readily recognizable by its very slender and threadlike form. Complete specimens are very difficult to obtain on account of the fragility of the worm; the head is usually broken off and lost. Several thousands of these worms have been found in a single chicken.

This tapeworm is one of the most common tapeworms of the duodenum of chickens and turkeys in the United States. Stafseth (1940) reported this species of tapeworm as a parasite of quail in Michigan. Ward (1946) listed *H. carioca* as a parasite of the quail in Mississippi.

**Life history.** Guberlet (1919) observed that chickens became infected with this tapeworm after they had been fed stable flies caught around poultry yards. It has been demonstrated by Jones (1929) and by Cram and Jones (1929) that dung beetles act as intermediate hosts.

Horsfall (1938) successfully grew cysticercoids of this species in *Tribolium castaneum* and *T. confusum.* When flour beetles containing cysticercoids of *H. carioca* were fed to young chickens, the latter became infected with the adults of this worm. Cysticercoids develop in beetles to a stage which is infective for chickens within approximately three weeks. Development of the adult worm in the chicken to the time when gravid segments are passed requires from two to four weeks.

**Pathology.** This tapeworm sometimes occurs in large numbers in chickens and turkeys; but it has very little, if any, effect on the growth rate of the birds, according to Luttermoser (1940).

*Hymenolepis cantaniana* (Polonio, 1860)

**Synonym.** *Taenia contaniana* Polonio, 1860.

**Description.** Mature specimens about 2 cm. long. Rostellum and suckers unarmed (Fig. 33.13 B). Genital pores unilateral, anterior to middle of segment margin. Testes 3 in number, usually arranged in a transverse row.

This species has been reported from poultry in the United States, Puerto Rico, Europe, and Asia. It is reported from quail collected in Maryland.

**Life history.** The development of the cysticercoid of this species of tapeworm is rather unique. As observed by Jones and Alicata (1935), the terminal buds arise from the many branched individual, and ultimately develop into infective larvae (Fig. 33.13 A). Dung beetles serve as intermediate hosts of this tapeworm. From two to three weeks are required for the bladderworm to develop into the adult tapeworm in the avian host.

**Pathology.** No definite pathological conditions have been associated with this species.



Fig. 33.13. *Hymenolepis contaniana.* A—developing larvae. B—head. Original.

*Hymenolepis tenuirostris* (Rudolphi, 1819)

**Synonyms.** *Taenia tenuirostris* Rudolphi, 1819; *Drepanidotaenia tenuirostris* (Rudolphi, 1819) Railliet, 1893.

**Description.** Mature worms 10 to 25 cm. long. Rostellum slender, with about 10 hooks, 20 to 23μ long (Fig. 33.14 A). Genital pores unilateral. Testes 3 in number, in a transverse row. Eggs (Fig. 33.14 B) not in capsules.

**Life history.** Unknown.

**Pathology.** Cram (1928) reported this parasite to be present in large numbers from the goose in Oregon, and regarded it as responsible for heavy losses. The affected birds showed symptoms of weakness, emaciation, incoordination, and diarrhea.

*Hymenolepis compressa* (Linton, 1892)

**Synonym.** *Taenia compressa* Linton, 1892.

**Description.** Mature worms up to 4 cm. long. Suckers unarmed (Fig. 33.15 A); rostellum with 10 hooks, 50 to 58μ long (Fig. 33.15 B and D). Testes 3 in number, in a more or less straight row across the segment (Fig. 33.15 C).

Sprehn (1932) listed this tapeworm as a parasite of ducks and geese from North America.

**Life history.** Unknown.

**Pathology.** Unknown.

*Hymenolepis coronula*
(Dujardin, 1845)

**Synonyms.** *Taenia coronula* Dujardin, 1845; *Weinlandia coronula* (Dujardin, 1845) Mayhew, 1925.

**Description.** Mature worms 1 to 2 cm. long. Suckers unarmed; rostellum armed with a crown of
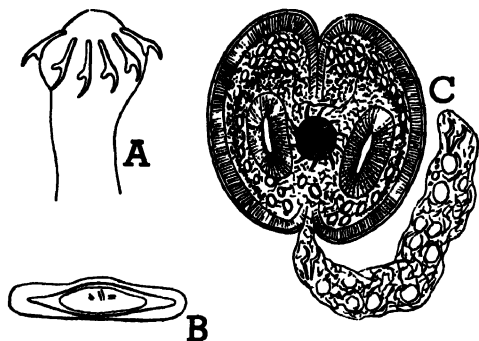
FIG. 33.14. *Hymenolepis tenuirostris.* A—head with rostellar hooks. B—egg. (From Krabbe, 1869.) C—cysticercoid. (From Hamann, 1889.)

18 to 26 hooks, 9 to 18μ long, with short handle and a strong guard which is almost as long as the blade (Fig. 33.16 A and B). Testes 3 in number (Fig. 33.16 C). Eggs not contained in capsules.

**Life history.** The eggs of this tapeworm are ingested by small crustaceans, the embryos hatching and developing to cysticercoids in the body cavity of these animals. When these infested crustaceans are swallowed by waterfowl, the cysticercoids develop to adult tapeworms in the intestines of the birds. Joyeux (1920) demonstrated that snails may carry cysticercoids of this species for a time after having eaten infested crustaceans. Birds may become infected by eating snails infested with cysticercoids.

**Pathology.** Pillers (1923) reported a heavy infestation with this species and with *H. megalops* and *Aploparaksis furcigera* as "apparently the cause of 'going light' and deaths" in ducks in England. Kingscote (1932) reported an enzootic in a flock of ducks in Canada caused by this species, the parasites being present in large numbers. Schofield (1932) reported heavy mortality among ducklings in Canada due to *H. coronula.*

*Hymenolepis lanceolata* (Bloch, 1782)

**Synonym.** *Taenia lanceolata* Bloch, 1782.

**Description.** Mature worms 3 to 13 cm. long. Segments 20 to 40 times as wide as long. Suckers unarmed; rostellum with 8 hooks, 31 to 35μ long, with handle longer than blade, and guard slightly salient (Fig. 33.17 C and B).
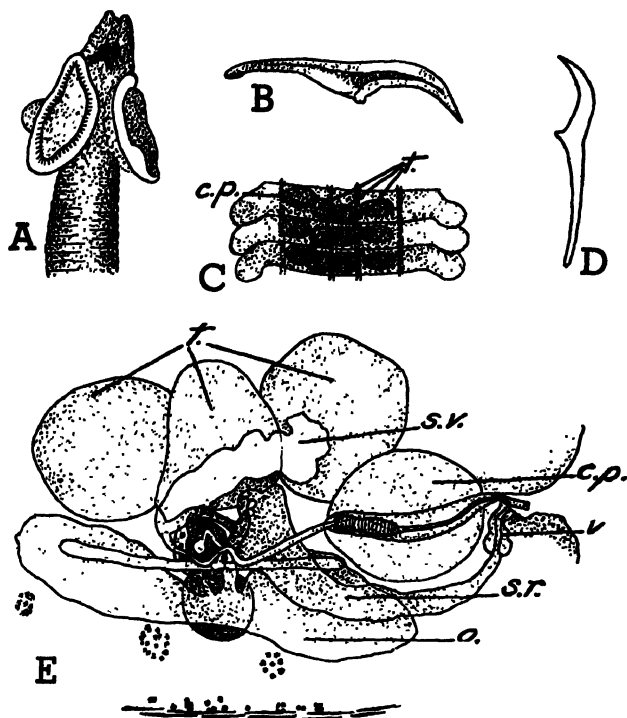
Fig. 33.15. *Hymenolepis compressa.* A—head. B—rostellar hook. (From Linton, 1892.) C—mature segments. D—rostellar hook. E—portion of transverse section through pore of mature segment (c.p.—cirrus pouch, o.—ovary, s.r.—seminal receptacles, s.v.—seminal vesicle, t.—testis, v.—vagina). (From Kowalewski, 1907.)
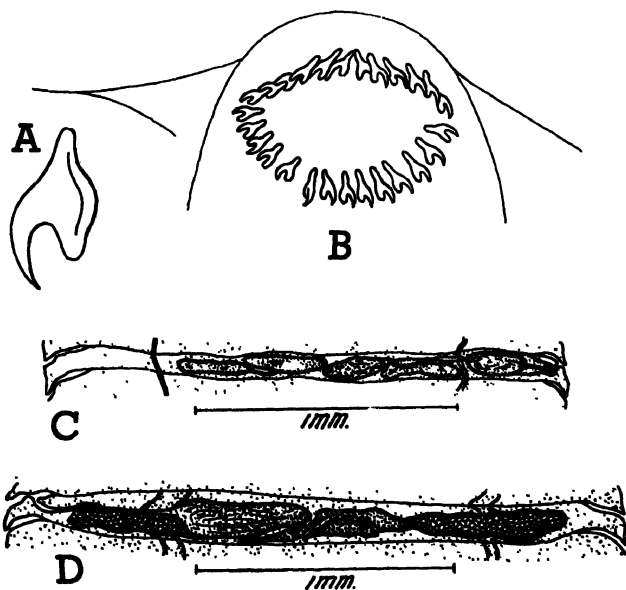


Fig. 33.16. *Hymenolepis coronula.* A—rostellar hook. B—hook crown in place. (From Krabbe, 1869.) C—mature segment with male genitalia. D—mature segment with female genitalia. (From Meggitt, 1920.)

Genital pore at anterior corner of segment margin, testes 3 in number, in a transverse row. Eggs not in capsules.

Quortrup and Shillinger (1941) reported *Hymenolepis* sp. (probably *H. lanceolata*) from the Canadian goose in Utah.

**Life history.** Ruszkowski (1932) demonstrated larvae (Fig. 33.17 D) of this species developed to the cysticercoid stage in small crustaceans (Fig.
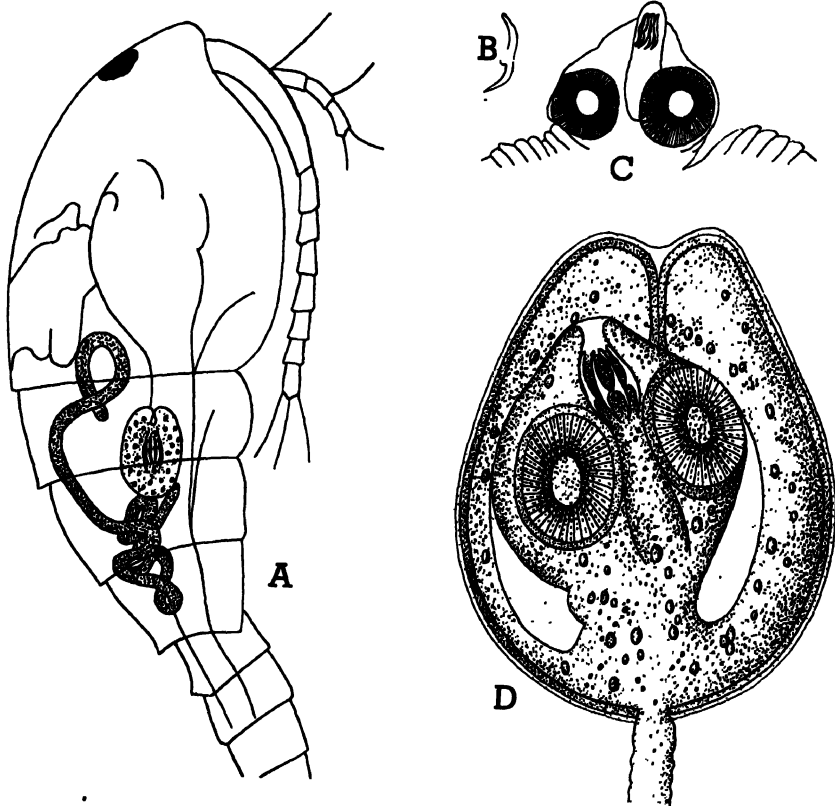


FIG. 33.17. *Hymenolepis lanceolata*. A—cysticercoid in *Diaptomus gracilis* showing long caudal appendage. B—rostellar hook. C—head. D—cysticercoid. (A and D from Ruszkowski, 1932; B and C from Southwell, 1930.)

33.17 A) in about six weeks at 9° to 12° C. The time required for the development of the adult worm in the primary host has not been determined.

**Pathology.** Emez (1929) described an epizootic, chiefly among young geese but also in some older birds. Muscular incoordination was the chief symptom. Post-mortem examination showed a catarrhal inflammation of the intestinal mucosa.

<center>*Hymenolepis megalops* Nitzsch, in Creplin, 1829</center>

**Synonyms.** *Taenia megalops* Nitzsch, in Creplin, 1829; *Weinlandia megalops* (Nitzsch, in Creplin, 1829) Mayhew, 1925.

**Description.** Mature worms 3 to 6 mm. long. Head very large, 1 to 2 mm. wide (Fig. 33.18). Suckers and rostellum unarmed. Testes 3 in number. Eggs not in capsules.

This tapeworm may be readily distinguished from other species found in poultry by its extraordinarily large head and its preference for the cloaca and bursa Fabricii. It has been found on a number of occasions in wild ducks.

Green (1938) reported this tapeworm from wild ducks in Minnesota. It has been collected on a number of occasions from wild ducks in Montana by Wehr.

**Life history.** Unknown.



FIG. 33.18. *Hymenolepis megalops*. Head. (From Stiles, 1896.)

**Pathology.** Pillers (1923) reported a heavy infestation with this worm and with *H. coronula* and *Aploparaksis furcigera* as "apparently the cause of 'going light' and of deaths in ducks in England."

*Hymenolepis tritesticulata* Fuhrmann, 1906

**Synonym.** *Weinlandia tritesticulata* (Fuhrmann, 1906).

**Description.** Mature worms 25 cm. long. Suckers unarmed; rostellum with 10 hooks, 32μ long (Fig. 33.19 B). Testes 3 in number. Eggs not in capsules.

This species of tapeworm has been reported by Linton (1927) as occurring in wild ducks of North America.

**Life history.** Unknown.

**Pathology.** Unknown.

*Hymenolepis introversa* (Mayhew, 1925)

**Description.** Mature worms 5 to 8 cm. long. Suckers unarmed (Fig. 33.20 A); rostellum armed with 20 hooks, 17 to 20μ long (Fig. 33.20 B). Genital pores in anterior region of right segment margins. Testes 3 in number, irregularly lobed.

This species of tapeworm has been reported by Mayhew (1925) as occurring in the duck from Illinois.

**Life history.** Unknown.

**Pathology.** Unknown.



FIG. 33.19. *Hymenolepis tritesticulata*. A—ovary and vitelline gland. B—rostellar hook. C—poral region showing part of cirrus pouch with internal sacculus accessorius. (From Fuhrmann, 1907.)

**SYMPTOMS**

Everything else being equal, the severity of the symptoms resulting from tapeworm infestations apparently depends to some extent on the number of worms present, on the diet, and on the age of the birds. Few, if any, clinical symptoms are observed in lightly infested birds. Heavily infested birds sometimes show marked retardation in growth rate.

Harwood and Luttermoser (1938) demonstrated experimentally that the growth rates of two- to four-week-old chicks fed an adequate diet and having infestations at autopsy ranging in numbers from 15 to 155 *Raillietina cesticillus* were definitely retarded. Ackert and Case (1938) reported weight retardation and reduced sugar and hemoglobin content of the blood in three- to four-month-old birds each having at autopsy infestations of 4 to 25 *Raillietina cesticillus*. Levine (1938) found that the difference between the mean weights of chickens experimentally infected with *Davainea proglottina* when seven weeks of age and held under observation for thirteen weeks was 12 per cent less than the controls. Alicata (1939) experimentally determined that birds receiving animal-protein supplements (fish meal and dry skim milk) had, at autopsy, an average of 14 tapeworms (*Hymenolepis exigua*) while a similar number of birds receiving plant-protein supplements (yeast, sesame meal, peanut oil, and soybean meal) had an average of 66 tapeworms. In contrast to the above observations, Luttermoser (1940) reported that the growth rates of twenty-two four-week-old Rhode Island Red chickens experimentally fed 1,000 cysticercoids of *Hymenolepis carioca* were practically the same as those of an equal number of controls held under similar conditions.



FIG. 33.20. *Hymenolepis introversa.* A—head. B—rostellar hook. (From Mayhew, 1925.)

Birds of all ages harbor tapeworms. However, Ackert and Reid (1937) have demonstrated that concomitant with an increase in age of the bird there is a corresponding increase in resistance to tapeworm infestation.

A number of clinical symptoms have been inadvertently ascribed to tapeworm infestations. At the present time there is not sufficient experimental evidence to show that such clinical symptoms as cyanosis, lameness, poor feathering, and failure to come into, or stay in production are due solely to the presence of these parasites.

## DIAGNOSIS

Diagnosis of tapeworm infestation by examination of the fresh droppings for the presence of eggs or segments is unreliable. Even in cases of heavy infestation, segments or eggs are sometimes absent. It has been shown by Harwood (1938) that segment production in the tapeworm *Raillietina cesticillus* occurs in cycles, segment production being marked at first by a period of intense segment elimination, alternating with periods in which no segments, or only a relatively few segments, were eliminated.

The diagnosis of poultry taeniasis is best made at necropsy. The intestine of the supposedly infested bird is slit open with an enterotome, spread out flat on the bottom of a suitable container and examined carefully for the white ribbon-like worms. If this method of examination reveals no worms, a small amount of water may be added to the container. The water will cause the worms, if present, to float to the surface or they may be seen swaying back and forth in the water above the opened intestine. In infestations involving tapeworms of the smaller species, the individual worms are often so small that they are overlooked. Therefore, examination under the binocular microscope of the intestinal scrapings is frequently necessary to detect such small species as *Davainea proglottina* and *Amoebotaenia sphenoides*.

## CONTROL OF POULTRY TAPEWORMS

**Prevention.** When one considers the number of tapeworms infesting poultry and their various intermediate hosts, the task of prevention of tapeworm infestation in birds raised under natural conditions seems impossible. Investigations have shown that many intermediate hosts of varying habits may serve experimentally as intermediate hosts of a single species of tapeworm. To prevent birds eating the many species of invertebrates, such as insects, snails, and slugs, is an inconceivable task.

The proper disposal of the droppings is unquestionably the most important single preventive measure for the control of tapeworm infestation. The droppings of infested birds are the source from which the intermediate hosts become infected. Therefore, care in removing the droppings frequently and disposing of them in such manner as to prevent the intermediate hosts from picking up the tapeworm eggs or gravid segments passed in the droppings is of primary importance. The body wastes from farm flocks can usually be disposed of by hauling them to the field and spreading thinly over the land. The action of the sun and wind will quickly dry out the droppings and destroy all parasitic material that may be present in them, since prolonged dessication is fatal to the infective stages of the parasites. This practice of disposing of poultry droppings not only serves to destroy parasitic material but also adds tremendously to the value of the land for growing crops. Poultry manure, when properly handled, is said to be an excellent fertilizer for garden and field crops. In order that the fertilizing value of poultry manure may not be lost, it must be stored in a suitable screened-in shed which has been provided with a cement floor and with a roof to keep out the rain and snow. The screens exclude the flying and crawling insects which may serve as intermediate hosts of poultry parasites.

The body wastes from backyard flocks usually must necessarily be handled in some other way, as accumulations from such flocks usually exceed the demands of the owner. In communities where large numbers of poultry raisers are within a short distance of each other, the droppings from their

birds are sometimes hauled to one or more centrally located storage sheds and retailed to the public at a reasonable price. This practice naturally raises the question as to how safe this manure is if used on land where other poultry are likely to run. Limited experimentation seems to indicate that much destruction of the tapeworm eggs may result from the self-sterilization process which takes place in stored manure.

Keeping the poultry runs clean and free of boards and rubbish of all kinds is important because such a practice eliminates the hiding places of many of the intermediate hosts.

## TREATMENT

Medication has served to reduce appreciably parasitism in many groups of livestock, but its applicability in the control of poultry parasites in general is limited.

A drug, in order to be a satisfactory poultry remedial agent, must be inexpensive, highly effective, nontoxic, and easy to administer. It is highly essential that a drug designed for the purpose of removing parasites from poultry possess the above qualifications, since the unit value of the domestic fowl is usually quite low.

A large number of drugs have been recommended for the removal of tapeworms from poultry, but none has proved to be satisfactory. For many years, kamala was highly recommended as a taenicide for poultry. Recent experiments, however, have shown this drug to exert only a "shearing effect," i.e., removing the strobilae but leaving the heads attached to the intestinal wall in an unharmed condition.

Reid (1940) found that starvation of birds infested with the cestode *Raillietina cesticillus* for 20 to 48 hours, including the over-night feeding intervals of the chicken, resulted in the loss of the strobilae (minus the head) of the worms. The loss of the tapeworm strobilae was apparently directly due to the partial starvation of the parasite, as it was determined that the glycogen store in worms from chickens starved 20 hours was lowered to less than one-twelfth of that found in tapeworms taken from non-starved birds. However, he (Reid, 1942) demonstrated that the tapeworm head was not affected by the long period of starvation. When normal feeding habits of the fowl were restored, new strobilae or segments were regenerated by the unaffected heads, and gravid segments appeared later in the feces of the birds. Therefore, it is obvious that the practice of starving tapeworm infested birds has an effect similar to that following the administration of kamala, i.e., a breaking off of the strobilae and leaving the heads attached to the mucosa. Because of its harmful effects on the health and growth of the birds and the rapid regeneration of new segments following the starvation period, such a procedure is not practical.

## REFERENCES

Ackert, J. E.: 1932. Fowl resistance to parasitism affected by vitamins A and B. Arch. Zool. Ital., Torino 16:1369.

—— and Case, A. A.: 1938. Effects of the tapeworm *Raillietina cesticillus* (Molin) on growing chickens. Jour. Parasit. 24:14.

—— and Reid, W. M.: 1937. Age resistance of chickens to the cestode *Raillietina cesticillus* (Molin). Jour. Parasit. 23:558.

Adams, F. M., and Geiser, S. W.: 1933. Helminth parasites of the chicken, *Gallus domesticus*, in Dallas County, Texas. Am. Midl. Nat. 14:251.

Alicata, J. E.: 1936. The amphipod. *Orchestia platensis* an intermediate host for *Hymenolepis exigua*, a tapeworm of chickens in Hawaii. Jour. Parasit. 22:515.

——: 1940. Poultry parasites. Annual Report, Hawaii Agr. Exper. Sta. (1939).

—— and Chang, E.: 1939. The life history of *Hymenolepis exigua*, cestode of poultry in Hawaii. Jour. Parasit. 25:121.

Cram, E. B.: 1928. The present status of our knowledge of poultry parasitism. No. Am. Vet. 9:43.

—— and Jones, M. F.: 1929. Observations on the life histories of *Raillietina cesticillus* and of *Hymenolepis carioca*, tapeworms of poultry and game birds. No. Am. Vet. 10:49.

Crawley, H.: 1922. *Davainea proglottina*, a pathogenic cestode, in American poultry. Jour. Am. Vet. Med. Assn. 61:305.

Emer, S.: 1929. (Cerebellar ataxia in geese as a result of infestation with *Hymenolepis lanceolata*.) (Russian text.) Vestnik Sovrem. Vet. Moskva, (94), Vol. 5 (21), Nov., p. 531.

Ferry, Q. B.: 1934. Studies on cestoda of poultry found in and around Douglas County, Kansas. Am. Midl. Nat. 15:586.

Gage, G. E., and Opperman, C. L.: 1909. Nodular taeniasis, or tapeworm disease, of fowls. Md. Agr. Exper. Sta., Bul. 139:73.

Grassi, B., and Rovelli, G.: 1899. Embryologische Forschungen an Cestoden. Zentralbl. f. Bakt. u. Parasitenk. 5:370 and 401.

Green, R. G., et al.: 1938. The occurrence of botulism in waterfowl in western Minnesota. Minn. Wildlife Dis. Invest. 3:128.

Guberlet, J. E.: 1919. On the life history of the chicken cestode, *Hymenolepis carioca* (Magalhães). Jour. Parasit. 6:35.

Harwood, P. D.: 1938. Reproductive cycles of *Raillietina cesticillus* of the fowl. Livro Jub. Lauro Travassos, p. 213. Rio de Janeiro, Institute Oswaldo Cruz.

—— and Luttermoser, G. W.: 1938. The influence of infections with the tapeworm, *Raillietina cesticillus*, on the growth of chickens. Proc. Helminth. Soc. Wash. 5:60.

Horsfall, M. W.: 1938. Meal beetles as intermediate hosts of poultry tapeworms. Poultry Sci. 17:8.

—— and Jones, M. F.: 1937. The life history of *Choanotaenia infundibulum*, a cestode parasitic in chickens. Jour. Parasit. 23:435.

Hudson, J. R.: 1934. Notes on some avian cestodes. Ann. and Mag. of Nat. Hist., Series 10 (80), 14:314.

Jones, M. F.: 1929. *Hister (Carcinops) 14-striatus* an intermediate host for *Hymenolepis carioca*. Jour. Parasit. 15:223.

——: 1930a. A new tapeworm from the guinea fowl, with cysticercoids in a ground beetle. Jour. Parasit. 16:158.

——: 1930b. Life history of *Metroliasthes lucida*, a tapeworm of the turkey. Jour. Parasit. 17:53.

——: 1936a. *Metroliasthes lucida*, a cestode of galliform birds, in arthropod and avian hosts. Proc. Helminth. Soc. Wash. 3:26.

——: 1936b. A new species of cestode, *Davainea meleagridis* (Davaineidae), from the turkey, with a key to species of Davainea from galliform birds. Proc. Helminth. Soc. Wash. 3:49.

—— and Alicata, J. E.: 1935. Development and morphology of the cestode, *Hymenolepis cantaniana* in coleopteran and avian hosts. Jour. Wash. Acad. Sci. 25:237.

—— and Horsfall, M. W.: 1935. Ants as intermediate hosts for two species of Raillietina parasitic in chickens. Jour. Parasit. 21:442.

Joyeux, C.: 1920. Cycle évolutif de quelques cestodes. Récherches expérimentales. Bul. de l'Inst. Pasteur 18:346.

—— and Baer, J. G.: 1937. Récherches sur l'évolution des cestodes de gallinacés. Compt. Rend. Acad. Sci. 205:751.

Kingscote, A. A.: 1932. Department of Parasitology. Rep. Ontario Vet. Coll. (1931) :60.

Levine, P. P.: 1938. The effect of infection with *Davainea proglottina* on the weights of growing chickens. Jour. Parasit. 24:550.

Linton, E.: 1927. Notes on cestode parasites of birds. Proc. U. S. Nat. Mus. (2656) 70, Art. 7, 73 pp. pls., 1–15, figs. 1–221.

Lopez-Neyra, C. R.: 1931. Revision del genero Davainea. Mem. Acad. Cien. Exact., Fes. y Nat. Madrid, s. Cien. Nat. 1:1.

Luttermoser, G. W.: 1940. The effect on the growth-rate of young chickens of infections of the tapeworm, *Hymenolepis carioca.* Proc. Helminth. Soc. Wash. 7:74.

Mayhew, R. L.: 1925. Studies on the avian species of the cestode family Hymenolepididae. Ill. Biol. Monogr. 10 (1), Jan., pp. 1–125, figs. 1–2, pls. 1–9, figs. 1–111.

Meggitt, F. J.: 1916. A contribution to the knowledge of the tapeworms of fowls and of sparrows. Parasitology 8:390.

————: 1926. The tapeworms of the domestic fowl. Jour. Burma Res. Soc., Rangoon 15:222.

Mönnig, H. O.: 1927. The anatomy and life history of the fowl tapeworm *Amoebotaenia sphenoides.* Union of So. Africa, Dept. of Agr., 11th and 12th Rep. Director Vet. Educ. and Res., Pt. 1:199.

Pillers, A. W. N.: 1923. Notes on parasites during 1922. Vet. Record 3:459.

Quortrup, E. R., and Shillinger, J. E.: 1941. 3,000 wild bird autopsies on western lake areas. Jour. Am. Vet. Med. Assn. 99:382.

Ransom, B. H.: 1905. The tapeworms of American chickens and turkeys. 21st Ann. Rep. Bur. An. Ind., U.S.D.A. (1904) :268.

Reid, W. M.: 1940. Some effects of short starvation periods upon the fowl cestode *Raillietina cesticillus* (Molin). Jour. Parasit. 26 (suppl.) :16.

————: 1942. The removal of the fowl tapeworm *Raillietina cesticillus* by short periods of starvation. Poultry Sci. 21:220.

Rietz, J. H.: 1930. Animal parasites of chickens in Ohio and West Virginia. Jour. Am. Vet. Med. Assn. 77:154.

Ruszkowski, J. S.: 1932. Cycle d'évolution du cestode *Drepanidotaenia lanceolata.* Bloch Acad. Polon. Sc. et Lett., Compt. Rend. Mens. Cl. Sc. Math. et Nat., Cracovie, (1), Jan., p. 4.

Schofield, F. W.: 1932. Heavy mortality among ducklings due to *Hymenolepis coronula.* Rep. Ontario Vet. Coll. (1931) :49.

Schwartz, B.: 1925. The chicken as a host for *Metroliasthes lucida.* Jour. Parasit. 12:112.

Southwell, T.: 1921. Cestodes from Indian poultry. Ann. Trop. Med. and Parasit. 15:161.

————: 1930. Cestoda. The Fauna of British India, Including Ceylon and Burma. 2:9.

Sprehn, C. E. W.: 1932. Lehrbuch der Helminthologie. Eine Naturgeschichte der in deutschen Saugetieren und Vogeln schmarotzenden Würmer, unter besonderer Berücksichtigung der Helminthen des Menschen, der Haustiere und wichtigsten Nutztiere. 998 pp., figs. 1–374. Berlin.

Stafseth, H. J.: 1940. Tapeworm infestation in poultry. Poultry Practice. A collection of discussions on poultry diseases and related subjects. Reprinted from Vet. Med. 34:763.

Stoddard, H. L.: 1931. The Bobwhite Quail, Its Habits, Preservation, and Increase. Charles Scribner's Sons, New York. 559 pp.

Ward, J. W.: 1946. A preliminary study of the occurrence of internal parasites of animals in Mississippi. Proc. Helminth. Soc. Wash. 13:12.

Wehr, E. E., and Coburn, D. R.: 1943. Some economically important parasites of the wild turkey and Hungarian partridge of Pennsylvania. Pa. Game News 13:14 and 31.

Williams, O. L.: 1931. Cestodes from the eastern wild turkey. Jour. Parasit. 18:14.

CHAPTER THIRTY-FOUR

# TREMATODES OF POULTRY

By EMMETT W. PRICE, *Zoological Division, Bureau of Animal Industry,*
*United States Department of Agriculture, Washington, D. C.*

☆    ☆    ☆

The trematodes or flukes are parasitic flatworms that as adults are devoid
of cilia or other locomotor appendages, but are provided with adhesive
organs in the form of suckers or other specialized structures.

The class **TREMATODA** is usually divided into two subclasses, namely,
the MONOGENEA and the DIGENEA. Some systematists recognize a
third subclass, ASPIDOGASTREA, which comprises a peculiar group of
flukes usually parasitic in bivalve mollusks. The MONOGENEA are para-
sites of cold-blooded animals, as a rule, and usually live on the exterior of the
body; they are peculiar forms having elaborate adhesive organs and direct
life histories. The DIGENEA are almost exclusively endoparasitic and for
the most part are provided with adhesive organs in the form of suckers; the
life histories are complex, involving alternation of generations and of hosts.

The digenetic trematodes are customarily divided into two orders, GAS-
TEROSTOMATA and PROSOSTOMATA. The GASTEROSTOMATA is a relatively
small group consisting of several genera characterized by having a single,
saclike digestive tract communicating with the exterior through a mouth
located near the middle of the ventral surface of the body. The gasterostomes
are parasites of fishes. The PROSOSTOMATA is the order to which all of the
poultry flukes belong. Members of this group are characterized by having
the mouth located at or near the anterior end of the body. The mouth is
usually surrounded by a sucker; a second sucker is usually present on the
ventral surface near the middle or, more rarely, at the posterior end of the
body.

**General morphology.** In general the body of the adult fluke is leaflike,
occasionally cylindrical, and frequently covered with scalelike spines. Except
for the blood flukes (Schistosomatidae), all trematodes of poultry are her-
maphroditic, that is, both the male and female organ systems are present in
a single individual. The male reproductive system usually consists of two
testes, vasa efferentia, a vas deferens which enlarges to form a seminal vesicle,
and a copulatory organ or cirrus surrounded by a saclike structure known
as the cirrus pouch. The female system consists of an ovary, vitelline or

yolk glands, an oötype or chamber in which the ovum and yolk cells are surrounded by shell material, and a long slender uterus the terminal portion of which is modified to form a vagina or metraterm. Both the male and female ducts usually open into a cavity or genital sinus which communicates with the exterior through the genital pore. In most flukes the genital pore is situated ventrally in the anterior part of the body. The digestive system is simple, and consists of a mouth, a short tube or prepharynx, a muscular bulb or pharynx, and a slender esophagus of varying length which branches to form the intestine; the intestinal branches are usually simple blind sacs or ceca, but in some forms the two branches are fused posteriorly (Cyclocoelidae) or united and terminating in a common cecum (Schistosomatidae). The nervous system consists of ganglia located in the pharyngeal region and of anteriorly and posteriorly directed nerves. The excretory system consists of an excretory pore that is located at the posterior end of the body, a bladder, two principal collecting ducts, and collecting tubules which ramify and terminate in flame cells.

**Development.** The developmental cycles of the trematodes of poultry are very complex. The eggs that are passed by the mature flukes customarily reach the exterior in the feces. On reaching water the eggs undergo embryonation and in the course of time hatch. The embryo or *miracidium* thus liberated swims about in search of a snail intermediate host. In some instances, as in the Cyclocoelidae and Schistosomatidae, the egg contains a fully formed miracidium at the time it is laid, and hatching takes place soon after it reaches water. In other instances, as in the Opisthorchiidae and Brachylaemidae, the egg contains at the time of deposition a fully formed miracidium which is not liberated until the egg is ingested by the snail host. On reaching a suitable location in the snail's tissues the miracidium is transformed into a *sporocyst*. When fully developed the sporocyst may give rise to a larva provided with a mouth and gut, which is known as a *redia*, and to *cercariae* (Echinostomatidae and Paramphistomidae), or it may give rise to *daughter sporocysts* and cercariae (Schistosomatidae and Strigeidae). The cercaria consists of a body, which becomes the mature fluke, and a tail which enables it to swim about. In some instances the cercaria may become encysted in the water or on various objects (Paramphistomidae and Notocotylidae) or penetrate into secondary intermediate hosts, such as snails, tadpoles, and fishes (Echinostomatidae, Opisthorchiidae, and Strigeidae), and become encysted. The encysted cercaria is known as a *metacercaria*. When the young encysted fluke, or metacercaria, is eaten by the final or definitive host the cyst wall is digested, and the young fluke is liberated and grows to maturity. In the Schistosomatidae the metacercarial stage is omitted; the cercaria penetrates the skin of the definitive host and on reaching the circulatory system develops into the adult fluke.

**Importance of flukes as parasites of poultry.** In comparison with the nematodes or roundworms of poultry, the flukes are of much less importance. In spite of the fact that a large number of trematodes are known from poultry—about fifty species from the chicken, twelve from the turkey, eight from the guinea fowl, two from the peafowl, twenty-eight from the pigeon, seventy-five from the duck, and twenty-four from the goose—only a few have been reported as causing serious injury. Many of the flukes of poultry have disease-producing potentialities, and serious losses may result if infestations are sufficiently large. In the case of the trematodes, as with other parasites, the amount of damage produced depends largely on the number of individuals harbored and to a lesser degree on the organs affected. In this chapter consideration is given mainly to those trematodes occurring in the United States which are actually or potentially capable of causing serious loss.

The species of flukes parasitizing poultry belong to eighteen families, the more important of which may be distinguished by the following key:

1. Sexes separate; parasites of the circulatory system . . Schistosomatidae
   Hermaphroditic; not parasites of the circulatory system . . . 2
2. Body fleshy, rounded or hemispherical; in cysts of skin . . .
   Troglotrematidae
   Body elongated, usually flattened; not in cysts . . . . . 3
3. Intestinal branches united posteriorly; parasites of respiratory system . . . . . . . . . . . . . Cyclocoelidae
   Intestinal branches not united posteriorly . . . . . . 4
4. With oral sucker only . . . . . . . . . . . 5
   With both oral and ventral suckers . . . . . . . . 6
5. Pharynx absent; uterus pretesticular; parasites of intestine and ceca . . . . . . . . . . . Notocotylidae
   Pharynx present; uterus largely post-testicular; parasites of kidney . . . . . . . . . . . . Eucotylidae
6. Acetabulum or ventral sucker located at posterior end of body . . . . . . . . . . Paramphistomidae
   Acetabulum or ventral sucker in middle, or anterior to middle, of body . . . . . . . . . . . . . . 7
7. Uterus passing between testes, reaching posterior end of body . . . . . . . . . . . Plagiorchiidae
   Uterus pretesticular . . . . . . . . . . . 8
8. Cirrus pouch absent; parasites of bile ducts . . . Opisthorchiidae
   Cirrus pouch present; not parasites of bile ducts . . . . . 9
9. Body divided by constriction into a cup-shaped anterior portion and a cylindrical posterior portion . . . . . . Strigeidae
   Body not divided as above . . . . . . . . . . 10

10. Gonads in posterior fourth of body; ovary between
    testes . . . . . . . . . . . . Brachylaemidae
    Gonads in middle, or posterior to middle of body; ovary in front of
    testes . . . . . . . . . . . . . . . 11
11. Oral sucker surrounded by an adoral disc armed with relatively
    large spines . . . . . . . . . . Echinostomatidae
    Oral sucker not surrounded by an adoral disc . . . . . . 12
12. Vitellaria tubular; eggs containing eye-spotted miracidia when
    deposited; parasites of the conjunctival sac . . Philophthalmidae
    Vitellaria follicular; eggs not containing eye-spotted miracidia
    when deposited; parasites of digestive tract . . . Psilostomidae

### TREMATODES OF THE SKIN

The skin fluke belongs to the Troglotrematidae. Members of this family have more or less plump, spiny bodies and frequently occur in cysts, usually in pairs.

### Collyriclum faba (Bremser, 1831)

**Synonym.** *Collyriclum colei* Ward, 1917.

**Description.** Body hemispherical, 4.2 to 8.6 mm. long by 4.5 to 5.5 mm. wide (Fig. 34.1). Oral sucker subterminal; acetabulum absent. Testes variable in shape; ovary T-shaped, with each of the branches divided into several lobes. Vitellaria in anterior part of body, somewhat asymmetrical, consisting of 6 to 9 groups of follicles on each side. Uterus greatly coiled, in posterior part of body. Eggs 19 to 21$\mu$ long by 9 to 11$\mu$ wide.

This parasite occurs encysted in the skin of chickens and turkeys and of a number of passerine birds. The cysts are 4 to 6 mm. in diameter, and each contains two flukes, one usually smaller than the other. An opening is present at the summit of the cyst through which the eggs of the flukes escape. In the United States this fluke has been reported from poultry in Minnesota where it was found in young chickens and turkeys by Riley and Kernkamp (1924). This parasite has also been reported by Marotel (1926) as parasitizing turkey poults in southeastern France.

**Life history.** The life history of this fluke is unknown. Like other members of the Troglotrematidae, this fluke undoubtedly requires a snail primary intermediate host and a secondary intermediate host, which is probably an arthropod. The theory of Jegen (1917) that infection is direct, since he reported that the eggs contained two embryos which were not miracidia but young flukes, is disproved by Tyzzer (1918), Riley and Kernkamp (1924), and Riley (1931) who observed miracidia escaping from the eggs, as in the case of other flukes. Riley is of the opinion that dragonfly larvae may serve as secondary intermediate hosts because the outbreaks of infestation among chickens and turkeys which he was able to observe occurred in birds having

access to wet or marshy places at a time in early summer when the dragonfly nymphs were emerging. Riley also "recovered from these nymphs metacercariae which suggest closely the characteristics of the adult Collyriclum."

**Pathology.** The encysted flukes are found mainly around the vent, but may occur elsewhere on the body of the affected bird (Fig. 34.2). In the cases studied in the United States there were no striking symptoms, but there is little doubt that extremely heavy infestations in young birds would prove fatal. In poultry of marketable age the presence of *Collyriclum* cysts would greatly decrease the value of the birds.

### TREMATODES OF THE EYE

The eye flukes belong to the family Philophthalmidae. They are relatively small trematodes with well-developed suckers and without spines; the gonads are in the posterior end of the body, the ovary being in front of the testes; the yolk glands or vitellaria are tubular. The best known species is *Philophthalmus gralli* Mathis and Leger, which has been reported from the chicken, peafowl, duck, and goose in Asia (Tonkin and Formosa). Several other species occur in poultry, including *P. anatinus* Sugimoto from the duck in China, *P. problematicus* Tubangui from the chicken and *P. rizalensis* Tubangui from the duck in the Philippines.



Fig. 34.1. *Collyriclum faba*. Ventral view. (From Kossack, 1911.)

The worms are attached by their suckers to the conjunctiva, causing congestion and erosion of the membrane. The conjunctival fluid contains blood, fluke eggs, and active miracidia. The life histories of the eye flukes are unknown. So far as known these parasites do not occur in American poultry.

### TREMATODES OF THE RESPIRATORY SYSTEM

Several species of the family Cyclocoelidae occur in domestic fowl. These flukes are relatively large and have flattened oval or lancet-shaped bodies. The oral sucker is weakly developed or absent, and the acetabulum or ventral sucker is absent or rudimentary. The digestive tract is continuous posteriorly. The gonads are in the posterior end of the body with the ovary variously arranged with respect to the testes.

*Typhlocoelum cucumerinum* (Rudolphi, 1809)

**Synonyms.** *Typhlocoelum flavum* (Mehlis, 1831) ; *T. obovale* Neumann, 1909.

**Description.** Body oval, 6 to 15 mm. long by 2 to 7 mm. wide, yellow in color. Mouth terminal, not surrounded by an oral sucker; acetabulum absent. Intestinal tract continuous posteriorly and provided with median

diverticula. Ovary and testes in posterior part of body, the latter deeply lobed. Uterus greatly convoluted, in median field. Eggs 154 to 180μ long by 85 to 90μ wide.

This fluke occurs in the trachea of wild waterfowl in the United States and in Europe; it has been reported from the domestic duck in South America.



FIG. 34.2. View of abdomen of turkey showing cysts of *Collyriclum faba*. (Kernkamp, Univ. of Minn.)

**Life history.** Incompletely known, probably similar to that of *T. cymbium*.

**Pathology.** This fluke has been reported by Magalhães (1899) in Brazil as the cause of suffocation promptly resulting in death of some domestic ducks; the flukes were present in large numbers in the trachea and bronchi.

### *Typhlocoelum cymbium* (Diesing, 1850)

**Synonym.** *Tracheophilus sisowi* Skrjabin, 1913.

**Description.** Body oval, 6 to 12 mm. long by 3 to 6 mm. wide (Fig. 34.3) and similar in appearance to *T. cucumerinum*, except that the testes are rounded instead of lobed. Eggs 122 to 154μ long by 63 to 81μ wide, containing miracidia at time of oviposition.

This species is not uncommon in wild waterfowl in various parts of the world, including the United States, and has been reported from the duck and goose. It occurs in the trachea, bronchi, air sacs, and infraorbital sinus.

**Life history.** The life history of this fluke has been ascertained by Szidat (1932) and by Stunkard (1934). The eggs, which contain miracidia when deposited, hatch on reaching water. The miracidium swims about and on coming in contact with suitable snails (*Menetus planorbis, Helisoma trivolvis, Planorbis corneus, Lymnaea palustris,* or *L. ovata*) penetrates the tissues and liberates a redia which is present in the body of the miracidium. The redia increases in size and gives rise to tailless cercariae which escape and become encysted in the vicinity of the redia. Birds become infected by eating the snails harboring the encysted cercariae.

**Pathology.** The presence of large numbers of these flukes in the larynx and trachea of birds causes death by suffocation. Light infestations may cause little or no injury.

Several other cyclocoelids have been reported as parasites of poultry, namely, *Cyclocoelum mutabile* (Zeder) from the goose and turkey in Europe, Asia, and South America; *C. japonicus* Kurisu from the chicken in Japan; and *Hyptiasmus tumidus* Kossack from the goose in Europe.

In addition to flukes of the family Cyclocoelidae, Price (1937) reported *Clinostomum attenuatum* Cort (Clinostomidae), normally a parasite of bitterns, from the trachea of a chicken in Nebraska. This was apparently a case of accidental parasitism acquired through the ingestion of a tadpole or young frog containing the larval fluke.
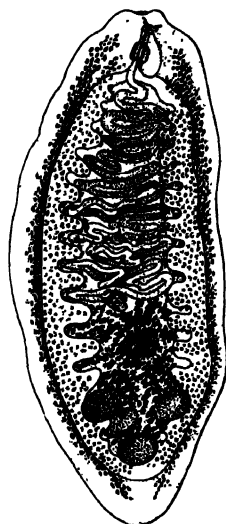


Fig. 34.3. *Typhlocoelum cymbium* (= *Tracheophilus sisowi*). Ventral view. (From Skrjabin, 1913.)

## TREMATODES OF THE DIGESTIVE SYSTEM

The digestive tract is a favorite location for flukes, and a large number of species representing many families have been reported from this organ system. Only a few of the more important of these species are discussed here.

### ECHINOSTOMATIDAE

Flukes of this family are characterized by having a kidney-shaped collar or adoral disc armed with one or two rows of spines.

### *Echinostoma revolutum* (Froelich, 1802)

**Synonyms.** *Echinostoma echinatum* (Zeder, 1803); *E. columbae* Zunker, 1925; *E. paraulum* Dietz, 1909; *E. miyagawai* Ishii, 1932; *E. cinetorchis* Ando and Ozaki, 1923.

**Description.** Body elongated, up to 22 mm. long (Fig. 34.4). Oral sucker surrounded by an adoral disc bearing 37 spines, 27 marginal and 5 on each ventral lobe. Acetabulum strongly developed, situated a short distance

posterior to oral sucker. Testes variable in shape, one behind the other; ovary pretesticular; uterus preovarial. Eggs 94 to 126µ long by 59 to 71µ wide.

This trematode occurs in the intestine, ceca, and cloaca of a wide variety of hosts, including the chicken, duck, goose, swan, turkey, pigeon, wild waterfowl, and some mammals. It has been reported under a variety of names from practically all parts of the world.

**Life history.** The life cycle of *Echinostoma revolutum* has been ascertained by Johnson (1920) and by Beaver (1937). The larval stages develop in fresh water mollusks of the genera Planorbis, Helisoma, Lymnaea, Stagnicola, and Pseudosuccinea. The cercariae which are formed in rediae and are provided with an adoral disc armed with spines as in the adult, escape and usually encyst in snails and tadpoles. The final hosts become infected through ingestion of the infested secondary intermediate hosts.

**Pathology.** In most instances and in light infestations, this fluke causes little injury. Heavy infestations in pigeons have been reported from Europe by Zunker (1925), Krause (1925), Bolle (1925), and van Heelsbergen (1927b), and in these cases losses have resulted. Zunker stated that the small intestine of the affected birds showed hemorrhagic inflammation, and similar findings were reported by Bolle. Krause collected about 5,000 echinostomes, apparently *E. revolutum,* from eight pigeons at Rostock; the more heavily parasitized birds died while the more lightly infested ones recovered after sickness lasting several weeks. In the cases studied by Bolle, there was a hemorrhagic diarrhea, and some of the pigeons died in an emaciated condition after being sick for 4 days. Krause reported as early symptoms the refusal of food, increased thirst, weakness in flight, and pronounced diarrhea; death occurred in 8 to 10 days following increased weakness. Van Heelsbergen in Holland also reported a severe enteritis in pigeons infested with an echinostome which appears to be *E. revolutum.* The birds showed atrophy of the pectoral muscles, engorged liver, and intestinal congestion, with the lumen of the gut filled with a hemorrhagic catarrhal secretion con-



FIG. 34.4. *Echinostoma revolutum.* Ventral view. (From Dietz, 1910.)

taining numerous flukes, 1,550 specimens being present in one pigeon. In the United States, Beaver (1937) reported a case of experimental infestation of a pigeon which developed a bloody diarrhea 10 days after infection. At necropsy 621 flukes were recovered, the majority being in the lower duodenum and upper ileum.

*Hypoderaeum conoideum* (Bloch, 1782)

**Synonyms.** *Echinostoma oxycephalum* (Rudolphi, 1819); *Opisthorchis pianae* Galli-Valerio, 1898; *Psilochasmus lecithosus* Otte, 1926.

**Description.** Similar in size and appearance to *Echinostoma revolutum* (Fig. 34.5). Adoral disc poorly developed, bearing a double row of 49 short spines. Eggs 95 to 108μ long by 61 to 68μ wide.

This species occurs in the small intestine of numerous wild waterfowl and has been found in the chicken, goose, and pigeon. It has been reported from the domestic duck in the United States by Stunkard and Dunihue (1931).

**Life history.** Similar to that of *E. revolutum*, snails of the genera Lymnaea, Stagnicola, and Planorbis serving as primary intermediate hosts and Planorbis and tadpoles serving as secondary intermediate hosts.

**Pathology.** Not well known. In a duck experimentally infected with 40 of these flukes, Vevers (1923) found a localized inflammation of the infested portion of the intestine, and the bird had been weak before death.

*Echinoparyphium recurvatum* (Linstow, 1873)

**Description.** Body (Fig. 34.6 A) 0.7 to 4.5 mm. long, with the anterior part strongly recurved ventrally. Adoral disc (Fig. 34.6 B) armed with 45 spines in a double row. General organization similar to that of *Echinostoma revolutum*. Eggs 108 to 120μ long by 64 to 84μ wide.

This parasite is widely distributed, having been reported from Europe, Asia, Africa, and North America. It occurs in various wild waterfowl and has been found in the duck, chicken, and pigeon in Europe, and in turkey poults in the United States.

**Life history.** Similar to that of *Echinostoma revolutum*. The larval
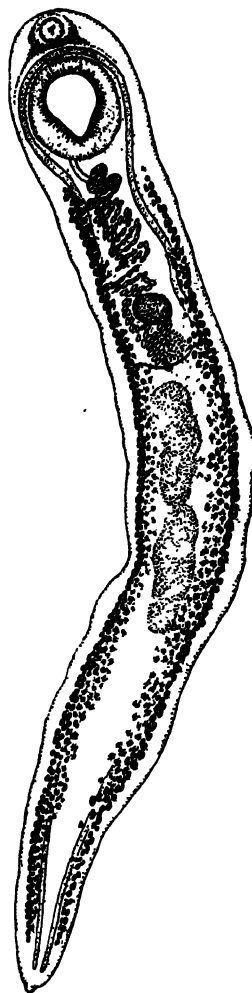


FIG. 34.5. *Hypoderaeum conoideum.* Ventral view. (From Dietz, 1910.)

stages develop in fresh water snails of the genera Lymnaea, Planorbis, and Viviparus. The cercariae encyst (Fig. 34.6 C) in snails and tadpoles. After ingestion of infested snails and tadpoles, the flukes become mature in the small intestine of the final host, and eggs appear in the feces in 5 to 7 days.

**Pathology.** Van Heelsbergen (1927a) reported that in Holland infested chickens showed a severe enteritis. The parasitized birds were emaciated, anemic, and developed weakness of the legs. Annereaux (1940) in California observed in a ten-week-old turkey with 267 adult flukes in the upper portion of the small intestine a "severe inflammation of the intestinal mucosa with cecal involvements consisting of a pasty, cheeselike mass which greatly distended the organs."
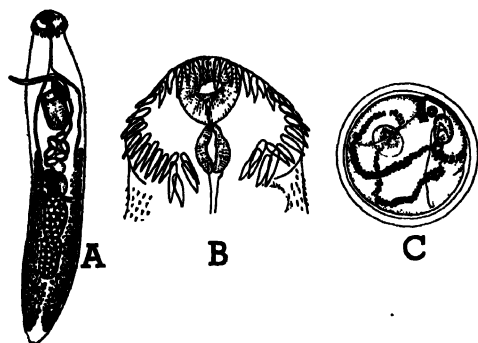


FIG. 34.6. *Echinoparyphium recurvatum.* A—entire worm, ventral view. B—anterior end. C—encysted cercaria. (From Bittner, 1925.)

#### PSILOSTOMIDAE

Flukes of this family resemble in general morphology species of the Echinostomatidae, but are not provided with a spine-bearing adoral disc.

### Ribeiroia ondatrae (Price, 1931)

**Synonym.** *Psilostomum ondatrae* Price, 1931.

**Description.** Elongate oval flukes measuring 1.6 to 3 mm. in length (Fig. 34.7); cuticle spiny. Oral sucker and acetabulum well developed. Esophagus with lateral diverticula. Testes in posterior end of body; ovary pretesticular. Vitellaria consisting of relatively large follicles extending from level of esophagus to posterior end of body. Uterus between ovary and acetabulum. Eggs 82 to 90µ long by 45 to 48µ wide.

This fluke, which was originally described from the muskrat in Canada by Price (1931a), occurs in the proventriculus of several fish-eating birds, including the California gull, osprey, and Cooper's hawk. It has also been reported in natural infestation in the chicken in Colorado by Newsom and Stout (1933), and in experimental infestations in the chicken, duck, pigeon, and canary by Beaver (1939).

**Life history.** Similar to that of the echinostomes. According to Beaver, the primary intermediate host is a fresh-water snail, *Helisoma antrosum percarinatum,* and the secondary intermediate hosts are fishes, including perch (*Perca flavescens*), rock bass (*Ambloplites rupestris*), smallmouth black bass (*Micropterus dolomieu*), pumpkin seed (*Eupomotis gibbosus*), bluegill (*Lepomis pallidus*), and bullhead (*Ameiurus*). The cercariae become encysted principally in the lateral line canal. In the final host the flukes reach maturity in 6 to 7 days.

**Pathology.** Newsom and Stout reported outbreaks of proventriculitis in two flocks of chickens in Colorado. The birds lost their appetite, stood around with their eyes closed and gradually wasted away. Gross examination "showed a very noticeable enlargement of the proventriculus. On opening this organ there seemed to be a deep reddening around the orfices of the . glands. In the more extreme cases there appeared to be a grayish exudate on the surface, simulating ulceration" (Fig. 34.8). Microscopic examination "showed that the surface of the mucous membrane was covered with a fibrinous exudate, the outer portion of which had become necrotic. Below this necrotic area was a thick zone heavily infiltrated with polymorphonuclear leukocytes. Under this, the mucous layer was quite edematous in which were scattered a few polymorphonuclear leukocytes and a few monocytes. In a few places small abscesses had formed in the lower portion of the mucous membrane." Beaver observed in experimental infestations in chickens and canaries that this fluke is fairly pathogenic, each worm forming in the proventriculus a separate lesion which is a deeply eroded pit with a raised orifice surrounded by a conspicuous reddish to purple area.

*Sphaeridiotrema globulus* (Rudolphi, 1814)

**Description.** Body piriform to globular, 0.5 to 0.85 mm. long (Fig. 34.9). Suckers well developed, acetabulum massive. Genital aperture lateral, at level of posterior margin of oral sucker. Testes in posterior end of body, one dorsal to other. Ovary pretesticular; vitellaria consisting of large follicles extending from intestinal bifurcation to level of anterior margins of testes; uterus relatively short, largely preacetabular. Eggs 90 to 105μ long by 60 to 67μ wide.



FIG. 34.7. *Ribeiroia ondatrae.* Ventral view. (Newsom, Colo. St. Coll.)

This fluke occurs in the small intestine and ceca of the wild duck in Europe and North America, and has been reported from the domestic duck and swan. In the United States this trematode was reported by Price (1934) as causing extensive loss among lesser scaup ducks near Washington, D. C., and it has also been found by Dr. J. N. Shaw in the domestic duck in Oregon.

**Life history.** As determined by Szidat (1937) in Germany, the cercaria of this fluke develops from redia in the snail *Bithynia tentaculata*. The cercariae become encysted between the shell and mantle of this snail, and birds acquire the parasite through ingestion of the infested mollusks, eggs appearing in the feces 5 to 6 days later.

**Pathology.** This fluke produces in wild ducks a severe ulcerative enteritis. In the cases studied by Price, the small intestine, especially the lower third,

showed marked congestion, hemorrhage, and ulceration, the lumen of the involved portion of the gut being filled with a cast composed largely of fibrin. Histologically the serosa, muscular, and mucous layers of the intestine showed evidence of acute hyperemia; the mucous membrane showed pronounced desquamation of epithelium, the villi being entirely denuded in most areas. There was severe ulceration in places, the ulcers frequently extending as deep as the muscularis and containing numerous flukes firmly attached by means of their powerful suckers. In the Oregon ducks the flukes were in the ceca where they caused extensive inflammation resulting in death of the affected birds.



FIG. 34.8. Proventriculus of chicken showing lesions caused by *Ribeiroia ondatrae*. (Newsom, Colo. St. Coll.)

### STRIGEIDAE

The strigeids are characterized by having the body divided by a constriction into two parts, an anterior cup-shaped portion containing the suckers and a peculiar tongue-shaped adhesive organ, and a cylindrical posterior portion containing the reproductive organs.

#### *Cotylurus flabelliformis* (Faust, 1917)

**Description.** Body 0.56 to 0.85 mm. long, anterior cup-shaped portion 0.20 to 0.28 mm. long, and posterior cylindrical portion 0.36 to 0.57 mm. long (Fig. 34.10). Genital aperture at posterior end of body. Eggs 100 to 112μ long by 68 to 76μ wide.

The fluke occurs in the intestine of a number of wild ducks in the United States, and has been reported from the domestic duck; it has also been reared experimentally in chickens.

**Life history.** The cercarial and precercarial stages occur in snails of the genera Helisoma, Planorbis, Stagnicola, Lymnaea, and Physa. The cercariae which develop in sporocysts in the snail are fork-tailed. The cercariae that escape from the primary snail intermediate host penetrate into other snails and develop into tetracotylid larvae. When snails containing the encysted tetracotylids are ingested by a definitive host, the worms mature in 3 to 4 days.

**Pathology.** According to Van Haitsma (1931), *C. flabelliformis* digests away the epithelium of the intestine of the host and causes a congestion of the subepithelial tissue. The symptoms showed by infested ducks appear to

vary greatly. Some of the infested ducks studied by Van Haitsma showed leg weakness, nervous twitchings of the head and wings, dyspnea, diarrhea, and irregular appetite, while others which had been given heavy doses of larvae died within a week without showing definite symptoms.

The only other strigeid reported in natural infestations from poultry in the United States is *Strigea falconis meleagris* Harwood (1931). This parasite was found in viscera of turkeys at Houston, Texas, and so far as known is of little economic importance.

A number of strigeid flukes have been reported from poultry in other parts of the world, and include *Strigea intermedia* Szidat from the duck and goose in Germany, and *Cotylurus cornutus* (Rudolphi) from the duck, goose, swan, and pigeon in Europe and South America.



Fig. 34.9. *Sphaeridiotrema globulus*. Ventral view. Original.

### BRACHYLAEMIDAE

Flukes of this family are characterized mainly by having the gonads in a linear series in the posterior end of the body, the ovary being situated between the testes. The genital pore is in the zone of the gonads.

### *Postharmostomum gallinum* (Witenberg, 1923)

**Synonyms.** *Harmostomum* (*Postharmostomum*) *horizawai* Ozaki, 1925; *H. annamense* Railliet, 1925; *H.* (*P.*) *hawaiiensis* Guberlet, 1928.



Fig. 34.10. *Cotylurus flabelliformis*. A—lateral view showing digestive and excretory systems. B—female genital system. C—male genital system. (From Van Haitsma, 1931.)

**Description.** Body linguiform, 3.5 to 7.4 mm. long (Fig. 34.11). Oral sucker and acetabulum relatively well developed, the latter situated about one-third of body length from anterior end. Intestinal ceca with wide serpentine undulations. Ovary between testes, in posterior end of body. Vitellaria lateral, extending anteriorly as far as posterior margin

of acetabulum; uterus extending anteriorly as far as intestinal bifurcation. Eggs 29 to 32μ long by 18μ wide.

This trematode occurs in the ceca of the chicken, turkey, guinea fowl, and pigeon in Europe, Asia, and Africa. It has also been reported from the chicken in Hawaii and Puerto Rico.



FIG. 34.11. *Posthar-mostomum gallinum.* Ventral view. (From Skrjabin, 1924.)

**Life history.** According to Alicata (1940), the eggs contain miracidia when oviposited. When these eggs are eaten by the snail *Eulota similaris,* the eggs hatch, and the miracidia enter the liver and develop into sporocysts. The cercariae developing in the sporocysts escape and leave the body of the snail; they may re-enter the same snail host or others of the same or a different species where they become encysted in the pericardial cavity. Another land snail, *Subulina octona,* has been shown to harbor the metacercariae, but it has not been determined whether this snail may also serve as a primary intermediate host. In the Orient, *Euhadra peliomphala, Philomycus bilineatus,* and *Eulota sieboldiana minor* have been reported as capable of serving as secondary intermediate hosts.

**Pathology.** So far as known, these flukes cause little or no injury to their bird hosts. It is possible that in extreme cases of heavy infestation, some irritation or inflammation of the ceca might result from the presence of the worms.

NOTOCOTYLIDAE

The notocotylids are small to medium-sized monostomes. The ventral surface is usually provided with rows of glands or ridges (absent in Paramonostomum). There is no pharynx, and the tips of the intestinal ceca pass between the testes which are located in the posterior part of the body. The eggs are small and are provided with a long slender filament at each pole.

*Notocotylus imbricatus* (Looss, 1893)

**Synonyms.** *Notocotylus seineti* Fuhrmann, 1919; *N. urbanensis* Harrah, 1922, in part; *N. intestinalis* Tubangui, 1932.

**Description.** Body elongate, oval, 2 to 4 mm. long (Fig. 34.12 A and B). Ventral surface with 3 linear rows of glands, 12 to 16 in median row and 12 to 17 in each lateral row. Eggs (Fig. 34.12 C) 17 to 20μ long by 9 to 12μ wide.

This species is perhaps the widest distributed of the notocotylids and occurs in Europe, Asia, and North America. It has been reported from ducks

and numerous wild waterfowl, and has been reared experimentally in the
chicken. According to Harwood (1939), this fluke has been collected from
domestic ducks in Oregon and New York.

**Life history.** The larval stages develop in the livers of snails of the genera
Bithynia, Lymnaea, and Physa. When the cercariae escape from the inter-
mediate host they encyst on the
shell of the snail or on other ob-
jects. When the cysts are in-
gested by suitable bird hosts, the
young flukes are liberated and
develop to maturity in the rec-
tum and ceca.

**Pathology.** Flukes of this
genus produce little injury to
their hosts. It is possible that if
present in large numbers, they
may cause some inflammation
of the rectum and ceca.

Other species of Notocotylus
reported from poultry are *N.
attenuatus* (Rudolphi) from
the duck, goose, turkey, and
chicken in Europe and Asia; *N.
ephemera* (Nitzsch) from the
chicken and duck in Europe;
*N. chionis* Baylis from the goose
in Europe; and *N. aegyptiacus*
Odhner from the duck in Africa (Egypt).



Fig. 34.12. *Notocotylus imbricatus* (=*N.
seineti*). A—ventral view, showing glands. B—
dorsal view, showing internal organization. C—
egg. (From Fuhrmann, 1919.)

*Catatropis verrucosa* (Froelich), a notocotylid having a glandular keel or
ridge instead of a median row of glands, occurs in the duck, goose, and
chicken in Europe. *Paramonostomum alveatum* (Mehlis) and *P. parvum*
Stunkard and Dunihue, species without ventral glands, occur in domestic
ducks in Europe and North America, respectively.

<center>PARAMPHISTOMIDAE</center>

This family comprises flukes having the acetabulum or ventral sucker
situated at the posterior end of the body. Only one species occurs in poultry.

<center>*Zygocotyle lunata* (Diesing, 1836)</center>

**Synonym.** *Zygocotyle ceratosa* Stunkard, 1916.

**Description.** Body ovate, up to 9 mm. long (Fig. 34.13). Oral sucker
subventral, provided with two evaginations or pouches. Acetabulum termi-
nal, large, with its posterior margin provided with a flap terminating on each

side in a conelike projection. Eggs 124 to 153μ long by 72 to 96μ wide.

This fluke occurs in the ceca of a number of wild waterfowl and has been reported by Price (1928) from the goose in the United States and by Caballero (1941) from the chicken in Mexico; it has also been reared experimentally in domestic ducks by Willey (1941).

**Life history.** The life history of this form has been studied in detail by Willey. The larval stages develop in the snail *Helisoma antrosum*. The cercariae which develop in rediae escape from the snail intermediate host and encyst on such objects as pond weeds and the shells of snails; infection of the final or definitive host occurs when the cysts are eaten. The flukes mature and give off eggs in about six weeks.

**Pathology.** So far as known, these flukes produce no appreciable injury to their bird hosts.



FIG. 34.13. *Zygocotyle lunata.* Ventral view. (Willey, New York Univ.)

### TREMATODES OF THE LIVER

The trematodes of the liver of poultry belong for the most part to the family Opisthorchiidae. They are semitransparent, usually elongate flukes, and occur in the bile ducts. Only one opisthorchiid has been reported as a parasite of poultry in the United States. This form, *Amphimerus* sp., was recorded by Price (1931b) from a turkey from North Dakota. The liver of this bird showed marked distention of the bile ducts and extensive pressure atrophy of the liver parenchyma.

Several opisthorchiids have been reported from ducks in various parts of the world, and include *Opisthorchis simulans* (Looss) from Europe; *O. longissimus* (Linstow) from Russia; and *Amphimerus anatis* (Yamaguti) from Japan. Closely related flukes of the genus Metorchis occur in ducks in Europe and elsewhere.

### TREMATODES OF THE URINARY SYSTEM

The trematodes occurring in the kidneys of poultry belong to the family Eucotylidae. These flukes lack a ventral sucker and a cirrus pouch, and have a long tortuous uterus filling the greater part of the pre- and post-testicular fields.

#### *Tamerlania bragai* dos Santos, 1934

**Description.** Body elongate, flat, up to 3 mm. long (Fig. 34.14). Oral sucker subterminal, acetabulum present, minute, according to Stunkard (1945). Pharynx relatively large; esophagus absent; intestinal ceca extending to and uniting near posterior end of body. Testes side by side in middle

of body; ovary more or less triangular immediately pretesticular. Vitellaria lateral, extending from level of pharynx to about one-fourth of body length from posterior end. Uterus convoluted, pre- and post-testicular. Eggs 31μ long by 13μ wide.

This fluke is found in the kidneys and ureters of pigeons in Brazil, Puerto Rico, and the Philippine Islands; it has also been reported from the chicken in Brazil.

**Life history.** Maldonado (1945) reported that the intermediate host is a land snail, *Subulina octona;* the larvae cycle is completed in about a month. Birds become infected upon ingestion of infected snails containing encysted metacercariae. Eggs are recoverable in the urine and excreta 23 days after infection.

**Pathology.** According to dos Santos (1934), the presence of the flukes in the kidney caused distention of the collecting tubules and a thickening of their walls, the lumen of the tubules being filled with amorphous and crystallized detritus. The parenchyma of the kidney showed extensive cellular infiltration, but the cortex was rarely involved. Maldonado and Hoffman (1941) noted similar changes in pigeons in Puerto Rico, but were of the opinion that the parasites caused no ill effect, since birds that were kept in cages for several months appeared unaffected by the parasites.

TREMATODES OF THE REPRODUCTIVE SYSTEM

The flukes of the reproductive system belong to the Plagiorchiidae, a family which is characterized by having the ascending and descending limbs



FIG. 34.14. *Tamerlania bragai.* Ventral view. (From dos Santos, 1934.)

of the uterus passing between the testes. Several representatives of this family occur in American poultry, the most important of which is discussed below.

*Prosthogonimus macrorchis* Macy, 1934

**Description.** Body piriform in outline, 5.26 to 7.56 mm. long (Fig. 34.15); cuticula spiny. Intestinal ceca simple, extending to near posterior end of body. Genital pore at anterior end of body, slightly to left of oral sucker; testes oval, opposite each other and about one-third of body length from posterior end. Ovary greatly lobulated, immediately posterior to acetabulum; vitellaria lateral, extending from acetabulum to testes; uterus with numerous coils in post-testicular part of the body. Eggs 28μ long by 16μ wide, with spine of variable shape and length at antopercular pole.

This fluke occurs in the bursa Fabricii and oviduct of the duck, chicken, and other birds in the United States; it is particularly common in the lake region of Michigan and Minnesota.

**Life history.** According to Macy (1934), "The sporocyst, found in the 'liver' of *Amnicola limosa porata*, produces the cercaria directly, there being no redia stage. The cercaria swims away from the snail host, and, if it is drawn into the anal opening of a suitable species of dragonfly naiad by the breathing movements of such a host, the tail of the cercaria is lost and the metacercaria thus formed makes its way to the muscle of the naiad, where it increases to about five times its original size. A thick wall with an outer radially-striated and an inner homogeneous layer (Fig. 34.16) now forms about the metacercaria, and the cyst usually comes to lie in the body cavity of the host. In the event the infested dragonfly naiad or adult is eaten by a suitable avian definitive host, the wall is digested off the cyst as it passes down the digestive tract of the bird. The worm then makes its way down the intestine to the cloaca and then to the bursa Fabricii or to the oviduct, where it develops into the mature trematode. Embryonated eggs produced by the fluke leave the host by way of the cloacal opening, and if they reach a lake inhabited by *Amnicola limosa* the latter become infested and sporocysts and cercariae develop." The important dragonfly hosts of *P. macrorchis* belong to the genera Leucorrhinia, Tetragoneuria, and Epicordulia.



FIG. 34.15. *Prosthogonimus macrorchis*. Complete worm from oviduct of chicken. (Macy, College of St. Thomas, St. Paul, Minn.)

**Pathology.** The lesions and symptoms caused by species of Prosthogonimus in Europe have been described by Hieronymi and Szidat (1921), Reinhardt (1922), Seifried (1923), de Blieck and van Heelsbergen (1922), and others, and in the United States by Kotlán and Chandler (1925) and Macy (1934). The disease in American fowl caused by *Prosthogonimus macrorchis* is essentially the same as that in Europe caused by *P. pellucidus*. Affected birds lose their normal activity and appetite, and there is a pronounced dropping off in egg production. The eggs that are produced frequently have very thin shells or no shells. On necropsy there may be extreme emaciation and anemia, and an adhesive peritonitis. The intestines may show pronounced hyperemia and be covered with a fibrinous exudate. The

oviduct may show similar changes, be distended, and contain considerable exudate and egg material. In some cases there may be a rupture of the oviduct and the secretions, albumen and yolk material, present in the body cavity. Flukes are present in the oviduct and in the egg material, as well as in the abdominal cavity in case of oviduct rupture. In some instances the peritonitis may be so pronounced as to be detected in the dead and unopened birds by the bluish-red color of the abdominal wall. These lesions, as well as the laying of thin-shelled eggs or of eggs with no shells, may be due to other causes, but it seems to be an established fact that the flukes may be a contributory cause, if not the actual cause, of this condition in many instances. At any rate, if such conditions are encountered in areas where dragonflies are breeding, such as in the lake regions of the country, prosthogonimiasis should be suspected.



FIG. 34.16. Section through metacercaria of *Prosthogonimus macrorchis* from abdomen of a dragonfly. (Macy, College of St. Thomas, St. Paul, Minn.)

## TREMATODES OF THE CIRCULATORY SYSTEM

All of the trematodes living in the circulatory system of birds belong to the family Schistosomatidae and are characterized by having the sexes separate. Several species, namely, *Bilharziella polonica* (Kowalewski), *Pseudobilharziella yokogawai* (Oiso), *Dendritobilharzia pulverulenta* (Braun), *Trichobilharzia ocellata* (La Valette), and *Gigantobilharzia monocotylea* Szidat occur in domestic waterfowl in Europe and elsewhere; but none of these is known to occur in this country. Several schistosomes are known from wild waterfowl in North America, and some of them will probably be found capable of infesting poultry. In spite of the fact that the blood flukes are serious parasites of man, those infesting poultry do not seem to cause comparable injury to their bird hosts. Szidat (1929) reported that in infestations with *Bilharziella polonica,* the eggs of the fluke in the intestinal wall caused slight connective tissue proliferation and some leukocytic infiltration; in infestations with *P. yokogawai,* Oiso (1927) noted pathological changes in the liver and intestine and arrested growth of the bird host.

## TREATMENT OF POULTRY TREMATODIASES

Owing to the fact that trematode infestations of poultry are rarely diagnosed ante mortem, practically nothing is known concerning effective treatment for their removal. Medicinal treatment would appear to be of no value

for the removal of the skin fluke, *Collyriclum faba,* surgical incision and mechanical removal of the worms seeming to be the rational procedure in such infestations.

Flukes occurring in the respiratory system, particularly of the nasal passages, trachea, and bronchi, might possibly be removed by inhalations of powdered drugs having vermicidal properties. The most promising of such drugs is barium antimonyl tartrate, which is highly effective against the poultry gapeworm. The method of administration of this drug is discussed on page 799.

For trematodes occurring in the digestive tract, carbon tetrachloride in doses of 1 to 3 cc., depending on the kind and size of the bird, might be tried. In case the flukes are in the proventriculus or in the upper part of the intestine, the drug may be introduced directly into the former organ by means of a syringe and rubber catheter. For flukes in the lower part of the intestine or in the ceca, 2 to 5 cc. of carbon tetrachloride in three to four times its volume of a bland oil, such as mineral oil or cottonseed oil, administered by rectal injection, would probably prove effective.

In infestations with the oviduct fluke, Prosthogonimus, carbon tetrachloride is again the most promising treatment. Schmid (1930) reported the administration to a hen of 1.5 cc. of this drug in an equal amount of flour paste. On the following day the bird received 1 cc. of the drug in 8 cc. of the paste, and on the third day 1.7 cc. of the drug in the same amount of the paste. At this time no more Prosthogonimus eggs could be found in the feces. On the day following the last treatment, a mass of egg yolk containing nine of the flukes was found in the cage. The bird was then killed and in the oviduct were found two small egg concretions in which several flukes were lying, and other flukes were imbedded in collections of mucus; all of the flukes apparently were dead. Other birds in the flock were treated, but the results were inconclusive.

No drug treatment of value is known for the destruction of fluke parasites of the excretory and circulatory systems.

### CONTROL OF POULTRY TREMATODES

In view of the fact that all of the trematode parasites of poultry require at least one snail intermediate host, measures for the prevention of fluke infestations must be directed toward control or eradication of these mollusks, or to keeping poultry away from areas where the parasites may be acquired. The latter is the easier and perhaps the most certain method of preventing the birds from acquiring trematode infestations, and consists of selecting areas for poultry raising that are as far removed as possible from streams or swampy places, or by fencing to keep the birds from ranging over such areas.

The control of the snail intermediate hosts may be accomplished either by draining the low, marshy places or by the use of chemicals that are toxic

to the snails. In the case of swampy areas drainage, either by means of open ditches or by the use of agricultural tile, will lower the water table to a point where there is insufficient surface moisture to enable the snails to propagate. If drainage should be too expensive or otherwise impractical, the snails may be destroyed by dusting the area with powdered copper sulfate or bluestone. The copper sulfate should be mixed with a carrier, such as fine sand or land plaster in the proportion of 1 part of the chemical to 4 to 8 parts of the carrier, and spread either by broadcasting by hand or by the use of hand or power dusters. For destroying snails in ponds and small lakes the powdered copper sulfate may be used as in the case of marshes. The chemical should be spread along the banks and in the water near the shore, as most of the snails will be found in these locations.

For destroying snails in streams, burlap sacks containing large crystals of copper sulfate may be placed in the streams at the uppermost part of the section to be treated in an amount sufficient to give a concentration of 1 part of the chemical to about 500,000 parts of water. The amount of the chemical necessary may be determined by ascertaining the cross-section area of the stream and multiplying by the velocity in order to get the flow in cubic feet per second. This result multiplied by 12, which is the amount in pounds of copper sulfate necessary to give a concentration of the chemical of 1 to 500,000 for a 24-hour period, equals the amount of copper sulfate needed for the treatment. For example, if a stream is 3 feet wide and 1 foot deep and the velocity is 2 feet per second, the flow is 6 cubic feet per second; this result times 12 equals 72 or the number of pounds of copper sulfate necessary for one treatment. This concentration of the chemical will kill most snails but is not injurious to livestock; it may kill some fish and will destroy algae and moss.

In some instances, especially with Prosthogonimus, where the fluke is acquired through the ingestion of dragonflies, keeping poultry away from the shores of ponds or lakes in the mornings when these insects are inactive is recommended.

## REFERENCES

Alicata, J. E.: 1940. The life cycle of *Postharmostomum gallinum*, the cecal fluke of poultry. Jour. Parasit. 26:135.

Annereaux, R. F.: 1940. A note on *Echinoparyphium recurvatum* (von Linstow) parasitic in California turkeys. Jour. Am. Vet. Med. Assn. 96:62.

Beaver, P. C.: 1937. Experimental studies on *Echinostoma revolutum* (Froelich), a fluke from birds and mammals. Ill. Biol. Monogr. 15, 96 pp.

———: 1939. The morphology and life history of *Psilostomum ondatrae* Price, 1931 (Trematoda: Psilostomidae). Jour. Parasit. 25:383.

Bolle, W.: 1925. Über einen Taubentrematoden aus der Gattung Echinostomum. Deut. tierärztl. Wochenschr. 33:529.

Caballero y. C., Eduardo: 1941. Parasitismo en *Gallus gallus* L. originado por *Zygocotyle lunatum* en la region de Lerma. III. An. Inst. Biol., Univ. Nac. Mexico 12:123.

de Blieck, L., and van Heelsbergen, T.: 1922. Trematoden als oorzaak van eileider-ontsteking en het leggen van windeieren. Tijdschr. Diergeneesk. 49:536.

dos Santos, V.: 1934. Monostomose renal dos aves domesticas. (Portuguese text; French and English summaries.) Rev. Dept. Nac. Prod. Animal 1:203.

Harwood, P. D.: 1931. *Strigea falconis meleagris*, n. var. Jour. Parasit. 18:51.

———: 1939. Notes on Tennessee helminths. IV. North American trematodes of the subfamily Notocotylinae. Jour. Tenn. Acad. Sci. 14:421.

van Heelsbergen, T.: 1927a. Echinostomiasis bij kippen door Echinoparyphium. Tijdschr. Diergeneesk. 54:413.

———: 1927b. Echinostomiasis bij de duif door Echinostoma. Tijdschr. Diergeneesk. 54:414.

Hieronymi, E., and Szidat, L.: 1921. Über eine neue Hühnerenzoötie, bedingt durch *Prosthogonimus intercalandus*, n. spec. Zentralbl. f. Bakt., I. Orig. 86:236.

Jegen, G.: 1917. *Collyriclum faba* (Bremser) Kossack. Ein Parasit der Singvögel, sein Bau und seine Lebensgeschichte. Zeitschr. wiss. Zool. 117:460.

Johnson, J. C.: 1920. The life cycle of *Echinostoma revolutum* (Froelich). Univ. Calif. Pub. in Zool. 19:335.

Kotlán, A., and Chandler, W. L.: 1925. A newly recognized fluke disease (prosthogonimiasis) of fowls in the United States. Jour. Am. Vet. Med. Assn. 67:756.

Krause, C.: 1925. Gehäuftes Sterben bei Tauben durch Echinostomiden. Berliner tierärztl. Wochenschr. 41:262.

Macy, R. W.: 1934. Studies on the taxonomy, morphology, and biology of *Prosthogonimus macrorchis* Macy, a common oviduct fluke of domestic fowls in North America. Univ. Minn. Agr. Exper. Sta., Tech. Bul. 98, 71 pp.

Magalhães, P. S.: 1899. Notes d'helminthologie brésilienne. 9. Monostomose suffocante des canards. Arch. Parasit. 2:258.

Maldonado, J. F.: 1945. The life cycle of *Tamerlania bragai*, Santos 1934, (Eucotylidae), a kidney fluke of domestic pigeons. Jour. Parasit. 31:306.

——— and Hoffman, W. A.: 1941. *Tamerlanea bragai*, a parasite of pigeons in Puerto Rico. Jour. Parasit. 27:91.

Marotel, G.: 1926. Une nouvelle maladie parasitaire, la monostomidose cutanée du dindon. Rev. vét. 78:725.

Newsom, I. E., and Stout, E. N.: 1933. Proventriculitis in chickens due to flukes. Vet. Med. 28:462.

Oiso, T.: 1927. On a new species of avian Schistosoma developing in the portal vein of the duck, and investigations of its life-history. (Japanese text; English summary.) Taiwan Igakkwai Zasshi. (270):848.

Price, E. W.: 1928. The host relationship of the trematode genus Zygocotyle. Jour. Agr. Res., U.S.D.A. 36:911.

———: 1931a. Four new species of trematode worms from the muskrat, *Ondatra zibethica*, with a key to the trematode parasites of the muskrat. Proc. U. S. Nat. Mus. (2870), 79, Art. 4:1–13.

———: 1931b. Trematode of genus Amphimerus in liver of domestic turkey. Jour. Parasit. 18:51.

———: 1934. Losses among wild ducks due to infestation with *Sphaeridiotrema globulus* (Rudolphi) (Trematoda; Psilostomidae). Proc. Helminth. Soc. Wash. 1:31.

———: 1937. A note on the occurrence of a trematode of the genus Clinostomum in a chicken. No. Am. Vet. 18 (April):33.

Reinhardt, R.: 1922. Seuchenhaft auftretende Eileiterentzündungen bei Hühnern durch Invasion von *Prosthogonimus intercalandus*. Berliner tierärztl. Wochenschr. 38:384.

Riley, W. A.: 1931. *Collyriclum faba* as a parasite of poultry. Poultry Sci. 10:204.

——— and Kernkamp, H. C. H.: 1924. Flukes of the genus Collyriclum as parasites of turkeys and chickens. Jour. Am. Vet. Med. Assn. 64:591.

Schmid, F.: 1930. Beitrag zur Geflügelparasiten-Behandlung. Tierärztl. Rundschau 36:313.

Seifried, O.: 1923. Durch Invasion von Trematoden (Prosthogonimus-Arten) verursachte seuchenhaft auftretende und tödlich verlaufende Eileiter-Erkrankungen bei Hühnern in Mecklenburg. Deut. tierärztl. Wochenschr. 31:541.

Stunkard, H. W.: 1934. The life history of *Typhlocoelum cymbium* (Diesing, 1850) Kossack, 1911 (Tremata, Cyclocoelidae). A contribution to the phylogeny of the monostomes. Bul. Soc. zool. France 59:447.

———: 1945. The morphology of *Tamerlania bragai* dos Santos, 1934. Jour. Parasit. 31:301.

——— and Dunihue, F. W.: 1931. Notes on the trematodes from a Long Island duck with description of a new species. Biol. Bul. 60:179.

Szidat, L.: 1929. Die Parasiten des Hausgeflügels. 3. *Bilharziella polonica* Kow., ein im Blut schmarotzender Trematode unserer Enten, seine Entwicklung und Uebertragung. Arch. Geflügelk. 3:78.

———: 1932. Zur Entwicklungsgeschichte der Cyclocoeliden. Der Lebenszyklus von *Tracheophilus sisowi* Skrj. 1923. Zool. Anz. 100:205.

———: 1937. Über die Entwicklungsgeschichte von *Sphaeridiotrema globulus* Rud. 1814 und die stellung der Psilostomidae Odhner im natürlichen System. I. Die Entwicklungsgeschichte von *Sphaeridiotrema globulus* Rud. Zeitschr. Parasitenk. 9:529.

Tyzzer, E. E.: 1918. A monostome of the genus Collyriclum occurring in the European sparrow, with observations on the development of the ovum. Jour. Med. Res. 38, n.s. 33:267.

Van Haitsma, J. P.: 1931. Studies on the trematode family Strigeidae (Holostomidae). No. XXII. *Cotylurus flabelliformis* (Faust) and its life-history. Pap. Mich. Acad. Sci. Arts and Letters. 13:447.

Vevers, G. M.: 1923. Observations on the life-histories of *Hypodaerium* [*sic*] *conoideum* (Bloch) and *Echinostomum revolutum* (Froel.): Trematode parasites of the domestic duck. Ann. Appl. Biol. 10:134.

Willey, C. H.: 1941. The life history and bionomics of the trematode, *Zygocotyle lunata* (Paramphistomidae). Zoologica 26:65.

Zunker, M.: 1925. *Echinostoma columbae* n. sp., ein neuer Parasit der Haustaube. Berliner tierärztl. Wochenschr. 41:483.

CHAPTER THIRTY-FIVE

## PROTOZOA

*By* E. R. BECKER, *Department of Zoology,*
*Iowa State College, Ames, Iowa*

☆   ☆   ☆

## COCCIDIOSIS OF THE CHICKEN

**Introduction.** Coccidiosis is a general term applied to infection with one or more of the many species belonging to the Coccidia, a subdivision of the great protozoan class **SPOROZOA,** all of whose representatives are parasitic and devoid of specialized organelles of locomotion in the vegetative stages. There are, however, as Tyzzer (1932) has emphasized, as many kinds of coccidiosis as there are species of coccidia, each with its characteristic symptoms. So far as fowl coccidiosis is concerned, all known types save one involve the digestive tract, whose cells are penetrated by the parasites. The minimal effect of the invasion is the destruction of a certain number of easily replaceable epithelial cells and mild intestinal catarrh, but in the more severe types there occur serious cell dislocations, inflammation, even hemorrhage, and sometimes death of the bird. The character and cause of the infection, while in the main attributable to the species of etiological agent, are affected also by the infective dosage, the susceptibility of the bird, and, perhaps, the nature of the bird's food.

While coccidiosis is cosmopolitan and occurs in practically all kinds of birds, the problem is simplified somewhat by the fact that the parasites are host-specific; that is, each species occurs in a single species of host or limited group of closely related hosts. In the latter case one particular species seems to be the optimum host for the parasite. However, a particular bird host may be suitable for the development of more than one species of coccidia. The common fowl harbors eight distinct species of the single genus Eimeria. Thus, while the problem of identifying species of coccidia is simplified by the host limitations of these parasites, it is, on the other hand, complicated by the possibility of occurrence of multiple species in a single host species.

As a group, the coccidioses of chickens are of more economic importance than those of any other domesticated bird. It is well known that under certain conditions extensive losses in poultry attributable to coccidia do occur, and are sometimes more far-reaching than at first realized. Turkeys, ducks, and guinea fowls certainly suffer less than do chickens from coccidial

infection, though it is believed that under certain conditions the infection may become serious in these birds. There are on record also disastrous outbreaks of renal coccidiosis in geese. Pheasants and quail, when raised in captivity, are said frequently to suffer serious losses.

**Taxonomic relationships.** Of the many known genera of the Coccidia there are but two of importance that the student of avian coccidiosis need keep in mind; namely, Eimeria and Isospora. They are readily distinguished on the basis of the development of their terminal stages, the oocysts, subsequent to passage by the host. The freshly passed oocysts of both genera consist of little more than a compound wall and a rounded mass of nucleated protoplasm separated by a jelly-like material. In the presence of moisture and oxygen there characteristically develop from the protoplasmic mass of Eimeria four spores, or sporocysts, each containing two more or less banana-shaped sporozoites (Fig. 35.1). The matured cyst of Isospora, by comparison, contains but two spores, each holding four sporozoites. The net result in the case of both genera is the production of eight sporozoites inside each oocyst. Incidentally, sporozoite formation is considered to represent the final phase of the life cycle.

The distribution of the two genera among the orders of birds has been best worked out by Boughton (1937a), Boughton, Boughton, and Volk (1938), and Boughton and Volk (1938) who report the occurrence of species of Eimeria in Pelecaniformes (pelicans, cormorants), Anseriformes (geese, ducks), Galliformes (chickens, grouse, pheasants, quail), Gruiformes (cranes, coots, moor-hens), Charadriiformes (plovers, sandpipers), and Columbiformes (pigeons, doves). Species of Isospora have been located in Falconiformes (hawks), Cuculiformes (cuckoos), Piciformes (woodpeckers), Strigiformes (owls), Coraciiformes (kingfishers), and Passeriformes (sparrows, robins, etc.). Both genera have been reported among the Charadriiformes. Our interest is limited mostly to the genus Eimeria, for it alone occurs in barnyard fowls and pigeons.

**The life cycle.** When a viable matured or sporulated oocyst of the genus Eimeria is ingested by a nonimmunized bird of a suitable species, eight sporozoites, whose development has been previously discussed, escape from the enclosing spore and oocyst in the intestine of the new host, and invade epithelial cells of the mucosa. Ordinarily the infective stages are ingested with food or drink, and the excystation process is facilitated by the body temperature of the bird and the action of digestive juices. Pratt (1937) observed excystation of *Eimeria tenella*, as indicated by liberated spores and sporozoites, in the crop of chicks as well as farther down in the intestine, as soon as 5 minutes after inoculation per os. Interesting at this point are Edgar and Herrick's (1944) findings that chicks infected with *E. tenella* in the morning when the crop is empty suffer more severely with the ensuing

disease than do birds infected later when the crop is full. Pratt noted that the spores passed through the side of the oocyst before the liberation of the sporozoites, the latter process altogether escaping his observation. By ligating the duodenum below the gizzard and introducing the sporulated oocysts into the duodenum with a hypodermic needle, he was able to demonstrate complete excystation in the duodenum without previous passage through the crop. His attempts to accomplish excystation in vitro were unsuccessful.



1—Micropyle.

2—Polar inclusion.

3—Oocyst jelly.

4—Endomembrane of cyst wall.

5—Middle or "granular layer" of cyst wall.

6—Exomembrane of cyst wall.

7—Sporocyst or envelope of a spore.

8—Oocystic or extra-residual body.

9—Stieda body of spore.

10—Refractile globule of sporozoite.

11—Sporozoite.

12—Sporocystic or intraresidual body.

13—Nucleus of sporozoite.

14—Small refractile globule at more attenuated end of sporozoite.

Fig. 35.1. Diagrammatic representation of a mature oocyst of the genus Eimeria (Becker).

Levine (1942a), however, failed to obtain infections in chicks inoculated with sporulated oocysts of E. tenella and E. necatrix when the pancreatic ducts were ligated, although he was successful in infecting birds with ligated pancreatic ducts with the merozoites of these species. He concluded that pancreatic juice is necessary for excystation of coccidia in chickens.

The presence of a sporozoite in the epithelial cell, or of the first generation trophozoite developing from it, is betrayed by an eosinophilic globule (an inclusion typical of sporozoites) observable in thin, stained sections. The young trophozoite, or schizont, is usually an ovoid or rounded body enclosing a nucleus in addition to the aforementioned globule. Growth of the schizont is accompanied by repeated binary divisions of the nuclear material so that it comes to possess a considerable number of nuclei by the time growth ceases (Fig. 35.2). The cytoplasm segments about the nuclei so that there are pro-

duced about as many first-generation merozoites as there were nuclei. Merozoites usually become sickle- or banana-shaped bodies usually pointed at the posterior end and, depending upon circumstances, either rounded or
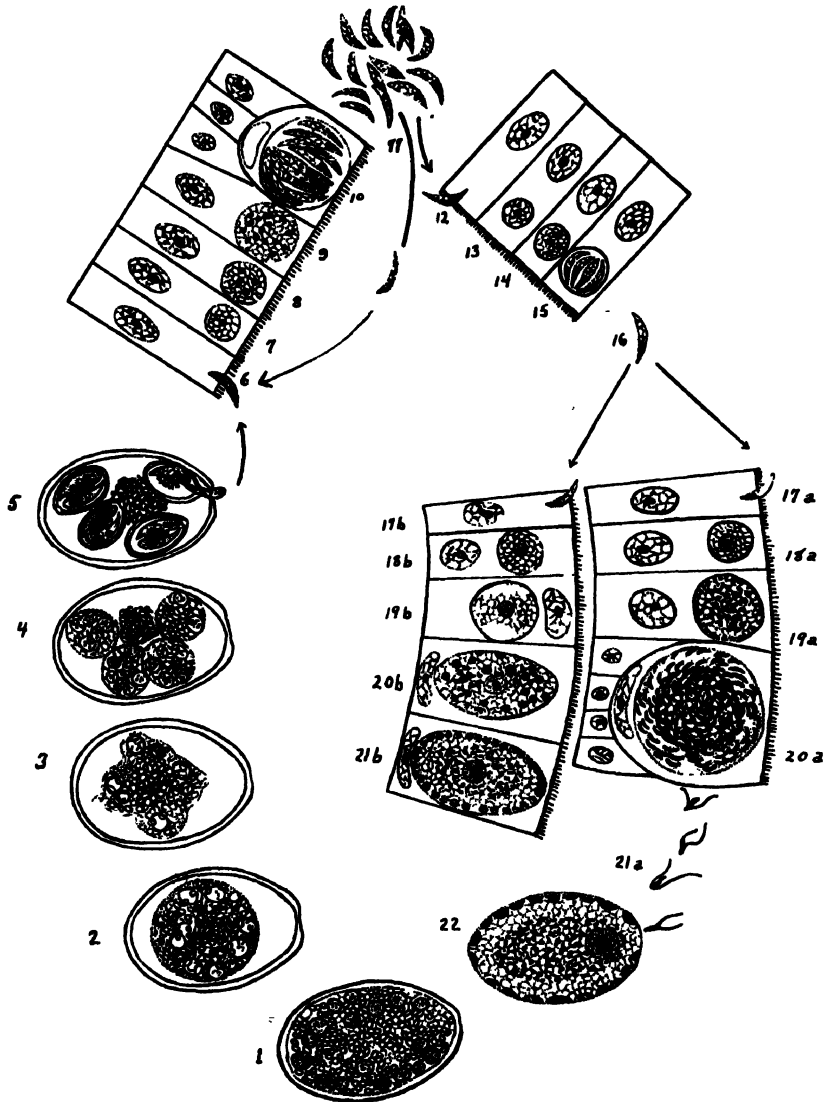


FIG. 35.2. Life cycle of *E. magna* (after Wasielewski). (From Becker.)

pointed at the anterior end. Before their release from the host cell they are recognizable as a clump with the individuals lying more or less parallel like the sections of an orange. The process just described, wherein a considerable number of merozoites are produced through asexual reproduction, is known

as schizogony. In most species of coccidia, as many of the first crop of mero-
zoites as can do so enter other epithelial cells, and the process is repeated in a
general way, but the eosinophilic globule does not occur in generations of
trophozoites subsequent to the first one. So far as is now definitely known,
the different generations of schizonts within any one species are character-
istic. They differ in size, shape, number of nuclei, and, hence, number of
merozoites produced, and in their effect upon the form, location, and activi-
ties of the parasitized host cell.

After several repetitions of the process of schizogony, there appears a
generation of merozoites that enter epithelial cells to develop into sexual
phases, or gametocytes. The latter are, in their younger stages, rounded
bodies, but as growth proceeds it becomes evident that they are of two sorts:
(1) males, or microgametocytes, in which growth is accompanied by many
nuclear divisions, and (2) females, or macrogametocytes, which also grow but
retain a single nucleus. From the microgametocyte there may develop as
many microgametes as nuclei, each of which is a minute flagellated body.
Fertilization is accomplished by the penetration of the microgamete into the
macrogamete (a matured macrogametocyte) through a micropyle. The re-
sulting zygote secretes a wall about itself, a process in which certain cyto-
plasmic granules seem to be involved, and it is known as an oocyst.

It is possible, however, that certain merozoites of the same crop that give
rise to the male and female gametocytes perpetuate the asexual cycle by giving
rise to another generation of schizonts, the merozoites from which in turn
again give rise to both merozoites and gametocytes, and so on. Such a con-
tinued procedure would be contingent, of course, on failure of immunity
to develop.

If there is a last generation of merozoites of which all survivors are fated
to become gametocytes, then oocyst production would wind up the cycle, and
an infected bird would cease to discharge oocysts soon after the intestinal
mucosa had become cleared of them, unless reinfection had taken place in
the meantime. In this case, it is customary to say that the cycle is "self-
limited." Although there are infections in which it seems that the cycle of
the parasite is self-limited (cf. Morehouse, 1938), it may indeed be otherwise
in the case of certain others. Thus, Boughton (1937b), after studying the
output of oocysts from English sparrows over periods of from two to five
months, concluded that Isospora infection in these birds is not self-limited,
but prolonged and chronic in nature.

**Periodicity.** Boughton in 1933 discovered diurnal gametic periodicity in
infections of the English sparrow with Isospora. The oocysts appeared in
the bird's droppings from 3 p.m. to 8 p.m. each day. They commenced to
appear in small numbers at the beginning of this period, reached a peak, and
declined again to small numbers at the end of the period. It was concluded

that the periodicity of oocyst production was affected, at least to a certain extent, by the metabolism of the host as it was regulated by the responses of the bird to light and darkness. A similar study (Boughton, 1937c) with pigeons infected with *E. labbeana* (supposedly) disclosed the peak of oocyst production at mid-day. Levine (1942a) studied the periodicity of oocyst discharge in *E. necatrix, E. hagani, E. maxima, E. mitis,* and *E. praecox* infections of chickens. In all cases except *E. necatrix* there was a tendency for the peak of oocyst discharge to occur during the 6 hours from 3 p.m. to 9 p.m., while in necatrix infections the highest oocyst elimination took place between 9 p.m. and 9 a.m. The latter phenomena may be due to the fact that necatrix oocysts develop in the ceca, which do not discharge their content regularly.

**Etiology.** There are at least eight valid species of coccidia known to occur naturally in chickens, if the rather recently described *Eimeria hagani* Levine (1938) and *E. brunetti* Levine (1942c) are included. Since it is practically impossible to present anything like a complete description of the eight species without involving the effect on the host, it is recommended that the section on pathogenicity be read in connection with this one.

Table 1, in large part reproduced from Becker (1934), shows that the oocysts of the eight species differ in respect to size, shape, and sporulation time (see Johnson, 1938, and Tyzzer, 1929). Nevertheless, it is most difficult to distinguish the species on the basis of oocyst characteristics alone, save that those of *E. maxima* can usually be readily identified by their larger size, together with their egg shape and rough walls. The time that intervenes between the feeding of sporulated oocysts and recovery of the next generation of oocysts in the feces (i.e., the prepatent period) varies from 4 days for *E. acervulina* and *E. praecox* and 5 days for *E. mitis* and *E. brunetti* to 7 days for *E. tenella, E. necatrix,* and *E. hagani.* Thus, oocysts recovered in the droppings before the middle of the sixth day would not be any of the three last-named species, and those of these species would not be expected to appear in a shorter interval. The region of the intestine parasitized, the position of the parasites in the intestinal mucosa, the macroscopic lesions, and the clinical type are also important characters that help in the identification of species. Tyzzer and Levine have also used the cross-immunity test to advantage.

Ordinarily it is difficult to identify the species on the basis of the morphology of the asexual stages in the intestinal wall when pathology and other characteristics are disregarded, but in the case of the *E. tenella* and *E. necatrix* there are certain stages that are indubitably peculiar to them. The second generation schizont of *E. tenella* commences its development in an epithelial cell of the cecal epithelium, but soon the parasitized cell grows, becomes rounded, and migrates into the underlying connective tissue. These large schizonts, measuring up to 54μ by 40μ, and subepithelial in position in the

cecal wall, are peculiar to *E. tenella*. *E. necatrix* produces second-generation schizonts similar to those of *E. tenella*, often even larger, but they are located in the subepithelial tissues of the small intestine. Oocysts in the cecal wall are indicative of the presence of either *E. tenella* or *E. necatrix* or of both.

**Transmission.** The only accepted natural method of transmission has been ingestion of the viable sporulated oocysts by a susceptible host. It is of interest, however, that several workers have succeeded experimentally in the transmission of infection by means of merozoites. Andrews (1927) first claimed such success in cats by intraduodenal injections of the merozoites of *Isospora felis*, but his account lacked assurance that the inoculum was entirely free from sporozoites. That Roudabush (1935) actually accomplished infection in the rat with merozoites of *Eimeria nieschulzi* ("*miyairii*") from a 5-day infection seems undeniable for he collected oocysts 2 days later. When the rat is infected with the sporulated oocysts of the same species, a full 7 days is required for the appearance of the oocysts. Krijgsman (1929c) states that Nöller succeeded in experimental transmission of coccidiosis to chicks through rectal injection of merozoite-containing material. Although Tyzzer (1929) had consistently failed to infect chicks with *Eimeria tenella* by cloacal injection of merozoites, Levine (1940c) accomplished this and also infected with merozoites injected directly into the small intestine through a catheter. He failed, however, in attempted gizzard infections. He likewise succeeded in attempted merozoite infections of the crop and intestine of chicks with *Eimeria maxima*, *E. praecox*, *E. necatrix*, and *E. hagani*. These successes encouraged Levine to express the opinion that under favorable conditions it is quite probable that merozoite infection occurs in the field. Certainly the chances of such transmission would be excellent in *Eimeria tenella* infection, which is characterized by copious bloody cecal evacuations charged with merozoites. Also, as Levine points out, cannibalism and devouring infected viscera from fresh carcasses would accomplish the same result.

The fact remains, however, that most infections have their incipiency in oocysts admitted into the digestive tract with food and drink, or by fouling of the beak while scratching litter or preening. In most poultry houses and runs where no special preventive measures are taken, there is ample opportunity for fecal material to lodge in wet drinking or feeding vessels and on damp litter or soil until sporulation has occurred. Under such conditions the whole flock is likely to become infected sooner or later, with the probability that certain birds will acquire much more massive infections than others. Andrews and Tsuchiya (1931), in a study of the distribution of oocysts on a Maryland poultry farm, found their greatest concentration to be in the vicinity of places where the birds spent the greatest percentage of their time; that is, roosts, brooding canopies, drinking fountains, and feed hoppers.

Dissemination of the oocysts is often more indirect. The hands, feet, and

## TABLE 1

### CHARACTERS FOR THE SEPARATION OF THE EIGHT SPECIES OF EIMERIA OCCURRING IN CHICKENS

| Characters | | E. tenella | E. mitis | E. acervulina | E. maxima | E. necatrix | E. praecox | E. hagani | E. brunetti |
|---|---|---|---|---|---|---|---|---|---|
| Oocyst | Size in $\mu$ | 26–19.5 X 22.8–16.5; av. 22.6X19 | 19.6–14.3X 17.0–13.0; av. 16.2X15.5 | 20.2–17.7X 16.3–13.7; av. 19.5X14.3 | 42.5–21.5X 29.8–16.5; av. 29.3X22.6 | 22.7–13.2X 18.3–11.3; av. 16.7X14.2 | 24.7–19.8X 19.8–15.7; av. 21.3X17.1 | 20.9–15.8X 19.5–14.3; av. 19.1X17.6 | 30.3–20.7X 24.2–18.1; av. 26.8X21.7 |
| | Shape | Broad ovoid | Subspherical | Egg-shaped | Egg-shaped | Oblong ovoidal | Ovoidal | Broadly ovoid | Egg-shaped |
| | Sporulation time | 48 hr. | 48 hr. | 21 hr. | 48 hr. | 48 hr. | 48 hr. | 24–48 hr. | 24–48 hr. |
| | Prepatent period | 7 days | 5 days | 4 days | 6 days | 7 days | 4 days | 7 days | 5 days |
| Region of intestine most heavily parasitized | | Schizonts and oocysts in ceca | Anterior small intestine | Anterior small intestine | Middle and posterior small intestine | Schizonts in small intestine, oocysts in ceca | Upper third of small intestine | Anterior half of small intestine | Posterior half of small intestine, rectum, ceca, and cloaca |
| Position of parasites in tissue | | Second generation schizonts subepithelial | Generally below nuclei of epithelial cells | Above nuclei of epithelial cells | Schizonts above nuclei. Gametocytes deep in epithelium | Similar to E. tenella | Below nuclei of epithelial cells | ? | Oocysts in all parts of cecal mucosa |
| Macroscopic lesions | | Hemorrhagic ceca | None | None | Exudate or flecks of blood on mucosa; intestinal wall thick | Whitish opacities and hemorrhage and exudate in small intestine | None, except mucous cast of intestine | Pin-head hemorrhages; severe catarrhal inflammation | Catarrhal enteritis with blood-tinged exudate |
| Degree of pathogenicity | | ++++ | + | + | ++ | ++++ | + | ++(?) | +++ |
| Development of immunity | | Sub-immediate | Delayed | Delayed? | Prompt | Delayed | Prompt | Prompt(?) | Sub-immediate |

utensils of the attendants undoubtedly serve mechanically to convey infec-
tion from one building or pen to another. Flies and fly larvae have also been
incriminated as mechanical vectors (Allen, 1932; Beaudette, 1928; Krijgs-
man, 1929a, b, and c), although Barger and Card (1935) remark that ade-
quate proof of fly carriage has not yet been brought forth. Metelkin (1935)
tested various species of wild and laboratory-bred muscoid flies, and found
them all capable of ingesting oocysts, which remained unaltered and viable
in the insect gut up to 24 hours, and in discharges until they dried. Dela-
plane and Stuart (1933) found that oocysts of avian coccidia in maggots
were destroyed or eliminated in the process of development of larvae into
adult flies. Less has been said about beetles, cockroaches, ants, and other
invertebrates, but these are also to be kept under suspicion as mechanical
vectors.

Birds and mammals visiting poultry runs probably carry oocysts about on
their feet, but definite proof is lacking. There is, however, definite proof
that oocysts, particularly the unsporulated, can pass through the intestines of
certain animals and remain viable. Pérard (1933), for example, proved that
dogs fed upon infected rabbit liver later egest the oocysts in such condition
that they are capable of sporulating and infecting susceptible rabbits. Rats
and mice may pass viable oocysts originating from other animals (Krijgsman,
1929b; Yakimoff and Iwanoff-Gobzem, 1931; Pérard, 1933).

It is sometimes stated that both diseased and resistant recovered animals
may act as contact carriers of coccidia. The statement is true, in a sense, but
it is probably also true that a chick may be very severely stricken with bloody
cecal coccidiosis on the fourth or fifth day of the infection, and die before
elimination of oocysts has commenced. In such a case the chick would not
yet have become a carrier unless, as was previously suggested, the infection
can be acquired through the infection of disseminated merozoites. In the
case of *Eimeria tenella* infection, oocyst elimination commences late on the
sixth day, and sometimes becomes so intense on the seventh or eighth day
that the cecal portions of the droppings consist of little else than oocysts and
a small amount of fluid. Usually the numbers passed decline rapidly there-
after, until after a week or two they are difficult to find in the droppings at all.
If, however, as often occurs in heavy infections, the cecal content commences
to caseate on the sixth or seventh day, there may be practically no oocysts
passed until days or weeks later when the cheesy core commences to liquefy.

Herrick, Ott, and Holmes (1936b) made a study of the length of time
chickens may serve as carriers of *Eimeria tenella* during a single infection.
Oocysts capable of sporulation and producing infections were found en-
meshed in the cecal wall 1½, 2, 2½, 3, 4, 4½, 5½, 6¾, and 7½ months fol-
lowing the infection date. In general, however, the longer the infection
persisted the fewer were the number of oocysts found, and the higher was

the percentage of them showing unmistakable signs of degeneration. After nine, ten, and twelve months the oocysts were not only rare, but totally incapable of sporulating or infecting. That there was a steady escape of oocysts was demonstrated by the occasional finding of oocysts in the cecal content up to 7½ months. Thus it appears that a recovered bird may serve as a carrier over a long duration of time.

Warner (1933) found soil previously seeded with the oocysts of chicken coccidia infective for 197 days, but not 217 and 231 days. Delaplane and Stuart (1935), working in Rhode Island, demonstrated that the oocysts of avian coccidia survived in soil from experimental ranges for four to nine months following the removal of chickens. In soil from a wooded range the oocysts remained viable at fifteen and eighteen months—a period of survival which appears phenomenal.

A number of years ago the suggestion was frequently made that eggs become contaminated by excreta containing coccidia as they are being laid, and that young chicks can become infected by ingesting shell during the hatching process. Johnson (1923) early minimized the importance of this proposed method of transmission on the basis of the susceptibility of oocysts to drying and certain other factors. Tyzzer (1932) commented upon the absence of coccidia in most of the day-old chicks on the market. Tyzzer, Theiler, and Jones (1932) actually smeared eggs with fecal material and potassium dichromate solution containing *Eimeria necatrix* and *E. praecox* oocysts, and incubated them. When the chicks began to pip the shell the fecal material was removed from the eggs and fed to susceptible chicks, but infection did not follow. Warner (1933) found that eggs dipped in suspensions of viable oocysts of poultry coccidia were not infective after 10 to 14 days incubation at 40–70 per cent relative humidity and 38–40° C. temperature. Ellis (1938), who made a more detailed study of *E. tenella,* found no viable sporulated oocysts on paper strips when they were kept at 45–70 per cent relative humidity and 100–104° F. for between 1 and 2 days, but at 91 to 93 per cent relative humidity and 100–104° F. they survived 3 and 4 days. Under conditions approximating those of normal egg incubation, sporulated oocysts did not live on the egg shell for more than a day or two. Furthermore, he found no evidence of coccidial infection in chicks hatched from eggs heavily smeared with oocyst-containing material and hatched in the regular manner. Thus, much of the available evidence points strongly to the effectiveness of the incubation process in destroying either sporulated or nonsporulated oocysts adhering to the egg shell.

Herrick (1935), however, appears to have been able to infect chicks by feeding egg shells intentionally contaminated with sporulated oocysts of *Eimeria tenella* and incubated for three weeks in an incubator kept at high relative humidity. The chicks that hatched from the eggs remained free from the infection.

**Host-specificity.** It is generally acknowledged that coccidia, particularly those of the common genus Eimeria, exhibit a marked degree of host-specificity. Evidence to support this will be found in many papers, some of the most pertinent of which are the following: Andrews (1927), Becker (1933), Corcuff (1928), Yakimoff and Iwanoff-Gobzem (1931), Yakimoff, Iwanoff-Gobzem, and Buewitsch (1932), Yakimoff and Gouseff (1933), Yakimoff, Iwanoff-Gobzem, and Matschoulsky (1936). The experiments reported in these papers involved many Eimeria species of both mammalian and avian origin and many mammalian and avian hosts, but few indeed were successful intraspecific cross-infections, some of which should be discussed.

Dieben (1924) succeeded in intraspecific cross-infections with *Eimeria miyairii* (=*E. nieschulzi*) between the closely related brown and black rats. Tyzzer and Jones (see Tyzzer, 1929) transferred *E. dispersa* from quail (bobwhite) to turkeys and occasionally to chickens, and *E. dispersa* from pheasants to quail, but second transfers in chickens and turkeys did not succeed. These failures were interpreted as indicating the slight adaptability of the quail coccidium to these hosts. Becker (1933) infected cottontails with *E. magna* from tame rabbits. It is to be noted that all of these successful attempts at cross-infection were between rather closely related groups.

The genus Isospora seems to show more laxity of host-specificity, at least among carnivores, for Andrews (1927) showed the *Isospora rivolta* and *I. felis* of the dog and cat are interchangeable between these two hosts. Furthermore, the dog has been infected by Balozet (1933) with a coccidium resembling *I. rivolta* from a mongoose, and by Yakimoff and Matikaschiwili (1932) with *Eimeria mephitidis* from a skunk. It is well known that Isospora is widely distributed in passerine birds, and lack of strict host-specificity is suspected to prevail there too, but critical experimental and taxonomic work bearing on this topic is still lacking, as was recently pointed out by Boughton.

There have been also certain claims of successful intraspecific infections that are open to serious question in view of lack of subsequent confirmation. Henry's (1931) claims for successful passages of *E. tenella, E. acervulina,* and an *E. mitis*-like coccidium (all taken originally from two species of California quail) to baby chicks, have been severely questioned by Tyzzer, Theiler, and Jones (1932) on the basis of certain possible deficiencies in her procedure. Tyzzer (1929) had failed to infect twelve chickens with *E. dispersa* from the quail, and Venard (1933) claims to have transmitted *E. tenella* of quail (bobwhite) origin to the chicken, although Patterson (1933) failed in infecting quail with *E. tenella, E. mitis, E. acervulina,* and *E. maxima* of chickens. Thus host-limitations of quail and chicken species require further investigation. The successful inoculation of chicks with Eimeria of rabbit origin was claimed by Uhlhorn (1926) and Krijgsman (1929c), and previously implied by Hadley (1910), but the attempted ex-

perimental transfers of Johnson (1923), Corcuff (1928), Tyzzer (1929), Yakimoff, Iwanoff-Gobzem (1931), and Crooks (1934) have disproved the likelihood that such cross-infection is possible. The writer knows of no proved instance of the infection of a bird with coccidia from a mammalian host or vice versa.

Many years ago wild birds, particularly the English sparrow, were blamed for the transmission of coccidiosis to fowls. Hadley (1910), who was confused concerning the etiological agent in blackhead of turkeys, discussed the English sparrow in terms that virtually condemned it as the source and disseminating agent in coccidiosis menacing the poultry-raising industry in all parts of the United States. He identified the coccidium of the sparrow with the "coccidium of blackhead in turkeys and of diarrhea in chicks." Smith and Smillie (1917) and Johnson (1923), however, pointed out that only two spores appeared in the developing sparrow coccidium, while there were four in those from domesticated fowls. While Hadley (1910) claimed to have infected fowls from sparrows and vice versa, Johnson (1923) could not corroborate these findings nor could he infect chicks with a coccidium from a blackbird, or vice versa. Boughton (1929) similarly found Minnesota sparrows infected with the two-spored type of oocyst (Isospora), but not the four-spored type of fowls (Eimeria), and he was unable to infect 3-day-old incubator chicks with the forms from sparrows.

Thus, the observed facts make it clear that, in general, coccidiosis in any particular species of bird or mammal is a problem more or less peculiar to it, and that "animal reservoirs" can usually be safely ruled out of consideration. Chickens acquire their infection from chickens, not from rabbits, rats, and mice, cattle, sheep, sparrows, or blackbirds. It is not to be implied, however, that animals which frequent poultry runs, such as rats, mice, and sparrows, cannot act as passive disseminators.

**Immunity.** Flocks may almost imperceptibly develop more or less protective immunity to coccidiosis by repeatedly picking up small amounts of infective material. It has been emphasized time and again that the ideal type of environmental control of coccidiosis is one which permits this immunizing process to proceed, rather than to attempt to maintain the flock coccidia-free. In the latter event a severe outbreak would follow accidental introduction of the infection. Immunity resulting from previous infections is the real reason older birds are more resistant to coccidiosis than younger ones. Immunization has also been produced by artificial inoculation with sporulated oocysts (Farr, 1943). (Cf. Becker, 1934, pp. 11–13, 40; Jankiewicz and Scofield, 1934.) Also, Herrick (1934) found that chicks displayed a natural resistance, and that chicks raised from parents that were particularly resistant to *E. tenella* were approximately 100 per cent more resistant than unselected chicks.

Various attempts have been made to attenuate the virulence of *Eimeria tenella* by treatment of the oocysts so as to effect immunity with a minimum of injury to the host. Jankiewicz and Scofield (1934) found that oocysts heated at 46° C. for 15 minutes before segmentation, and then fed to chickens after sporulation, conferred resistance to later inoculation with unheated oocysts and a minimum of injury to the host. One procedure that protected chicks against reinfection, and hence against the usual symptoms of coccidiosis, consisted of an initial inoculation of 649 such heat-treated oocysts, a second dose of 1,100, and a third of 3,300. The doses came at 5-day intervals. Heat treatment after sporulation was nearly as effective. Unheated oocysts kept at room temperature remained virulent. Waxler (1941b) X-rayed the oocysts with 9,000r. Such oocysts, when fed to 35-day-old chicks, caused some drop in hemoglobin concentration but no deaths. The mild infection conferred almost as much resistance as a severe attack resulting from untreated oocysts. Research on attenuation of coccidia for immunization purposes is still in the experimental stage.

**Pathogenicity.** The clarification of the problem of species identity in coccidial infections of fowls, principally through the work of Tyzzer and of Johnson, makes it possible to discuss pathogenicity much more authoritatively than was formerly possible before the etiology of the eight types of infection was understood. The pathogenicity problem, however, is by no means so simple that it may be discussed under eight headings, for the picture is complicated by the various forms assumed by infections with simple etiology depending upon size of infective dosage, host-predisposition, etc., and by special manifestations peculiar to mixed infections.

Tyzzer, Theiler, and Jones (1932) state that in common poultry certain species of coccidia are practically innocuous, while others are capable of producing serious and destructive outbreaks of disease. The same authors declare that their "consistent failure to demonstrate any pronounced pathogenicity on the part of *E. acervulina*" led them to abandon their former view and to consider the pathogenicity of the latter species unproved. Likewise they state that *E. praecox* "elicits no appreciable inflammatory reaction even in heavy infections, so that it may be regarded as practically innocuous as far as direct injury to the tissue is concerned." Tyzzer (1929, p. 309) had previously stated that *E. mitis* was not known to be of pathological significance. Thus three of the six or seven species of coccidia in chickens are not characterized by severe adverse effects on their hosts, according to Tyzzer, although Dickinson (1939) and Becker (1940) have presented definite evidence for a certain degree of pathogenicity on the part of *E. acervulina*, and Allen (1933) for *E. mitis*.

The pre-eminently pathogenic species are *E. tenella*, which so attacks the cecal wall as to produce an acute, hemorrhagic type of disease, *E. necatrix*.

which attacks the small intestine, so as to produce either an acute initial attack resulting in early death or a lingering illness characterized by progressive emaciation and general unthriftiness, and *E. brunetti,* which distributes itself in the mucosa of the lower half of the small intestine, rectum, ceca, and cloaca, causing more or less continuous, light daily losses in the flock, but leaving the birds in normal flesh. *E. maxima,* according to Tyzzer, attacks the middle and lower small intestine so that the wall becomes edematous and thickened, and covered on the mucosal side with a hemorrhagic exudate. *E. hagani,* according to Levine, causes a catarrhal enteritis in the anterior half of the small intestine.

**Specific pathological peculiarities.** *Eimeria tenella* is the cause of so-called cecal or bloody coccidiosis of chicks (Figs. 35.3 and 35.4). Involvement of the ceca rather than of the small intestine is one of its characteristic features. The severity of this type of coccidiosis is attributable to the second generation schizont, which causes infected epithelial cells to increase tremendously in size and assume a migratory habit. Through pressure or otherwise there is produced sufficient degeneration of the blood vessels and surrounding tissues to result in bleeding into the ceca, and the copious bloody discharges from the ceca. The discharges usually commence to appear bloody sometime before the end of the fifth day after infection, but a certain wateriness of the droppings is sometimes noted much earlier.

Oocysts appear in the droppings commencing on the seventh day. Their discharge continues more or less continuously over a considerable period. Herrick, Ott, and Holmes (1936b) found them in the droppings of chickens up to 7½ months after the infection date. Their study showed that following infection the oocysts are enmeshed in the tissue of the ceca where they remain viable for at least 7½ months. Sporulation of oocysts of *E. tenella* requires a little less than 48 hours.

*Eimeria necatrix* is another of the so-called pathogenic coccidia of the chicken. This species attacks the small intestine, with the maximum involvement near the middle. The second generation schizonts are similar in form and behavior to those of *E. tenella,* and like them produce the injurious effects. On the fourth and fifth days aggregations of these schizonts appear as small whitish opacities. Later, punctate hemorrhages appear in the center of the whitish areas, and may become so extensive as to obscure them altogether. As Tyzzer and collaborators state, "The unopened intestine thus presents a spotted appearance, the small whitish areas being intermingled with rounded, bright or dull red blotches of various sizes while transversely extending reddish streaks represent hemorrhages along the superficial vessels." There is profuse hemorrhage into the intestinal lumen.

Disease produced by *E. necatrix* may be of two types—acute or chronic. The former may result in the death of the bird 5 to 7 days after infection,

· while in the latter case the bird may linger on for a long time with a wasting illness. During the acute attack blood may be observed in the droppings.

*Eimeria necatrix* is unique among fowl coccidia in that, while the first two generations of schizonts develop in the small intestine, the merozoites
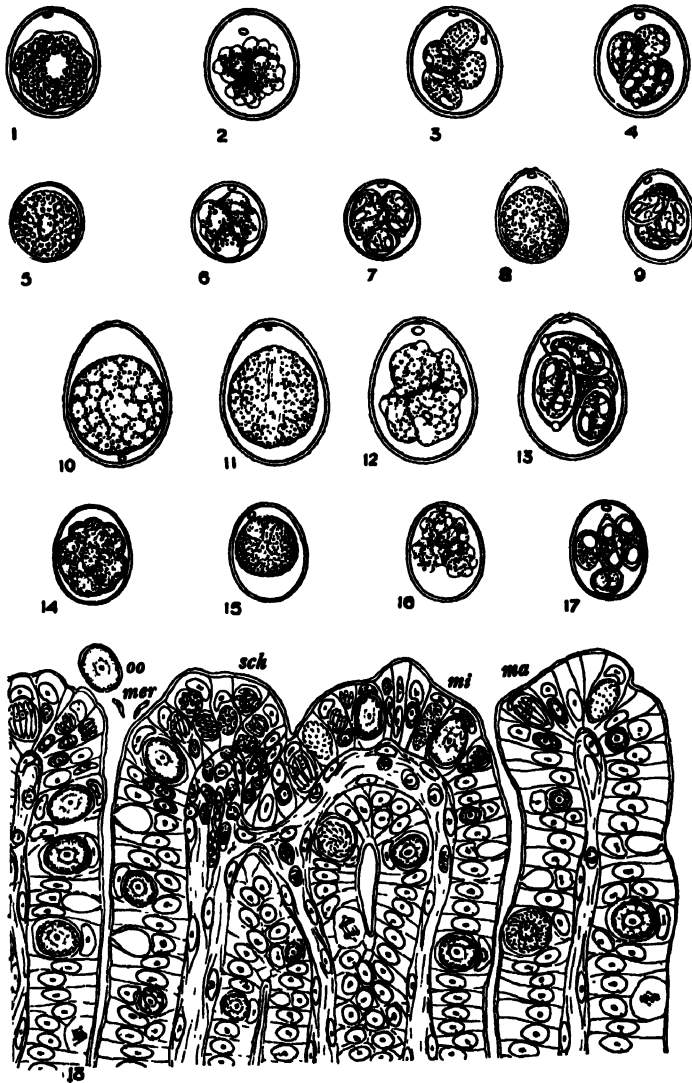


FIG. 35.3. Five species of Eimeria found in chickens. 1–17—×670. 1–4—stages in development of oocysts of *E. tenella*. 5–7—same for *E. mitis*. 8–9—same for *E. acervulina*. 10–13—same for *E. maxima*. 14–17—same for *E. necatrix*. 18—developmental stages in cecal epithelium from 7 to 9 days after infection. *oo*—oocyst. *sch*—third generation schizont. *mer*—third generation merozoite. *mi*—microgametocyte. *ma*—macrogametocyte. (All after Tyzzer, reproduced with permission of the Am. Jour. of Hyg.)

generated by the second generation schizonts migrate to the ceca where they invade the epithelium and develop, some into further generations of schizonts and some directly into oocysts.

Oocysts appear in the droppings on the seventh day after infection, and ordinarily require 2 days to sporulate. Tyzzer found that far more of them
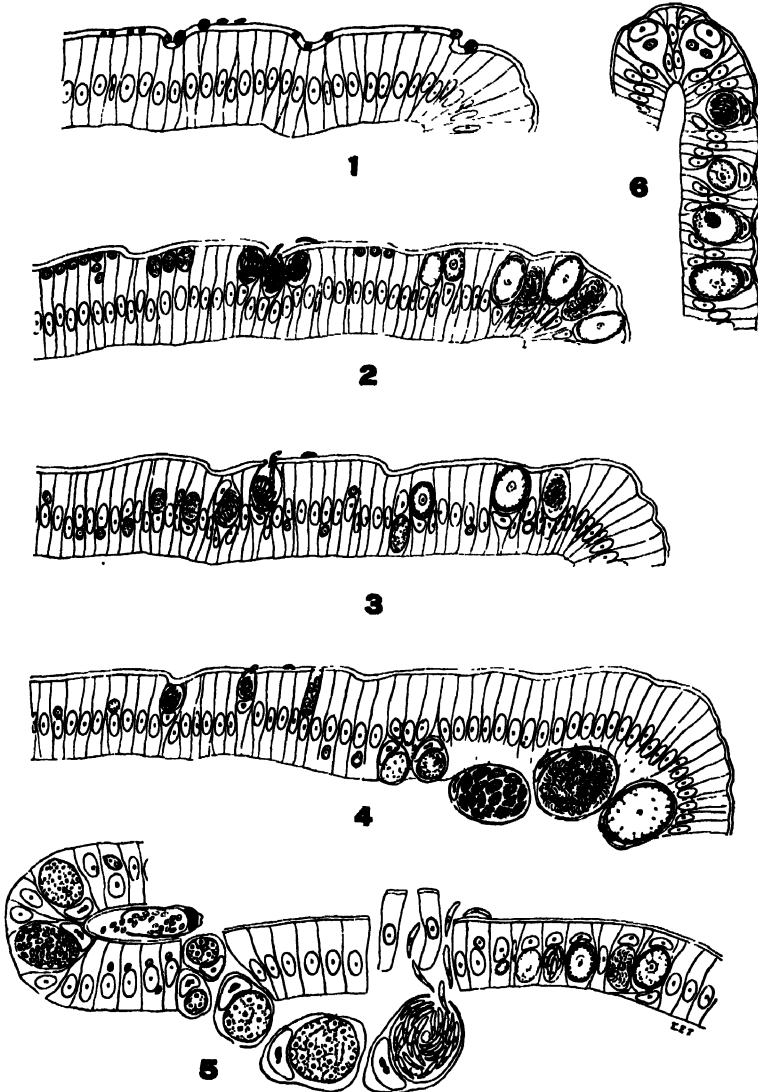


FIG. 35.4. Diagram illustrating the situation of different species of coccidia in the intestine of fowls and the reaction of the parasitized epithelium. 1—*Cryptosporidium parvum*. 2—*Eimeria acervulina*. 3—*E. mitis*. 4—*E. maxima*. 5—*E. tenella*. 6—*E. phasiani* (in the pheasant). (All after Tyzzer, reproduced with permission of the Am. Jour. of Hyg.)

are produced in light infections than in heavy ones. An infected bird may discharge oocysts over a prolonged period.

*Eimeria maxima* is the third of the definitely pathogenic species in the chicken, though far less lethal than the other two. Clinically, the recognition signs are dilation of the small intestine and thickening of the wall. The serous surface may show faint hemorrhages. The content is not bloody, but takes the form of viscid mucus, grayish, brownish, or pinkish in color. In some instances the portion of the feces from the small intestine may show flecks of blood. Infections sometimes terminate fatally, though the writer once infected a lot of thirty three-week-old birds without losses, or even serious symptoms.

Oocysts appear in the droppings on the sixth day after infection and continue for only a few days. After oocyst elimination ceases, the bird usually possesses a high degree of immunity to reinfection, and is definitely protected from recurring clinical symptoms. The oocysts of this species are quite characteristic, being the largest of all occurring in chickens (about 29.3μ by 22.6μ on an average), and having a slightly roughened wall.

*Eimeria acervulina* is, fortunately, not a severe pathogen, though it is perhaps the commonest of all the poultry coccidia. It is characterized clinically by numerous gray or whitish patches in the upper half of the small intestine, visible through the serous surface. These patches are caused by forming oocysts.

That the species is not very pathogenic is quite likely a fair statement of the situation. The writer once inoculated sixty white leghorn chicks with 300,000 sporulated oocysts each. The droppings which fell through hardware cloth onto newspapers were definitely more watery on the third day under the infected birds than under the controls, and the appetite was impaired for 2 or 3 days, but there were no losses. Oocysts are eliminated in tremendous numbers, commencing the fourth day.

*Eimeria mitis* grows in the small intestine throughout its entire length, but is most concentrated in the upper half. It is definitely not a serious pathogen. Using the test alluded to in the discussion of *E. acervulina*, that is, size of the wet rings around the portions of droppings of small intestinal origin, the writer has not been able to ascribe any edema of the small intestine to this species. The oocysts are small, with a tendency to the spherical, and usually are not abundant in the droppings.

*Eimeria praecox* develops in the upper third of the small intestine. This species is probably no more pathogenic than *E. mitis,* as the author has verified in an extensive test. Oocysts, however, are passed in abundance, commencing on the fourth day.

*Eimeria hagani* was recently established as a new species by Levine (1938). It will be described here in detail since it does not appear in either

Becker (1934) or Johnson (1938). The infection is characterized postmortem by numerous round hemorrhagic spots of the size of a pin head visible through the serosa in the duodenum and the upper half of the remainder of the small intestine, and comparatively few such lesions in the lower half. The oocysts, which develop mostly in the affected regions mentioned, commence to be discharged toward the end of the sixth day and appear in great numbers on the seventh day. Chickens immunized to *E. acervulina*, *E. praecox*, *E. mitis*, and *E. maxima*, respectively, can be infected with *E. hagani*, thus demonstrating the specificity of that form. Birds autopsied on the sixth day of the infection exhibit "a severe catarrhal inflammation of the duodenum and anterior half of the small intestine."

Levine's (1942c) account of *E. brunetti* indicates that the various stages of the parasite are distributed throughout the mucosa of the posterior half of the small intestine, rectum, ceca, and cloaca, and also the upper portion of the small intestine in heavy infections. In moderate infections there is a thickening of the gut wall, a pinkish or blood-tinged catarrhal exudate, and there may be also in the mucosa short, transverse red streaks, a millimeter or so in length, arranged in ladder-like fashion in long rows down the lower intestine and rectum. In severe infections there is an extensive coagulation necrosis and sloughing throughout the entire intestinal mucosa. Caseous cores may be found plugging the narrow portion of the ceca, but the dilated portions of the cecal wall are only moderately affected.

Ray (1945) has described *Wenyonella gallinae* from 4- to 6-week-old chickens in India. Sporulated oocysts of the genus Wenyonella, like Eimeria, contain four spores, and each of the latter, like Isospora, contains four sporozoites. The oval oocysts presented a punctate surface, rough in optical section, and measured $29.5\mu–33.5\mu \times 19.8\mu–22.8\mu$. The characteristically flask-shaped sporocysts measured $18.8\mu \times 8.0\mu$. At 28° C. in 2.5 per cent potassium dichromate solution, sporulation required 4 to 6 days. The infection was characterized by (1) blackish-green, semisolid excreta and intestinal content containing numerous oocysts and (2) pin-point hemorrhages in the mucosa and thickening and congestion of the terminal part of the intestine.

**Histopathology.** We are indebted largely to Tyzzer (1929), Tyzzer, Theiler, and Jones (1932), and Mayhew (1937) for knowledge concerning the histopathology in coccidiosis of the chicken. Since *E. tenella* is probably by far the most important of the severe pathogens, its effect on the tissues deserves special comment.

The first penetration of the host tissue is the entrance of the sporozoites into the basal portion of the cells lining the fundus of the cecal gland, where it grows into a schizont distinguishable by an eosinophilic globule persisting from the sporozoite. It increases in size, develops about 900 merozoites, and moves out through the distal end of the cell into the lumen, pushing the host-

cell nucleus before it. The merozoites are liberated, penetrate into other normal cells in the vicinity, and establish themselves in cells in a position distal to the nucleus, where they increase in size tremendously, causing the host-cell to become rounded and to assume amoeboid or wandering habits. They migrate into the tunica propria, or even the submucosa, where they increase in size to such an extent that by virtue of volume and numbers the cecal wall becomes congested, blood vessels become disrupted, and leakage of blood ensues. Thus is explained the hemorrhage that sometimes commences late on the fourth day of a heavy attack, and persists through to the sixth day. According to Tyzzer the bird may literally bleed to death.

Mayhew (1937) found that in light attacks severe damage to the tissue is but local and any destroyed epithelial or underlying tissues are regenerated. If there has been severe bleeding, a core will form in the lumen. In heavy attacks, on the other hand, a considerable area of the mucosa and submucosa may become congested, and the developing parasites may cause such disintegration of the tissue elements that the layers of the mucosa and submucosa lose their identity. There is such profuse discharge of blood cells, lymph, parasites, and tissue cells into the lumen of the cecum as to form a clot or core fitting the form of the ceca. The core to the cecal wall is adherent at first, but in a few days the surface liquefies so as to free it. It may be passed with the feces in the course of time, though sometimes the cores are retained. Allen (1934) and Mayhew (1937) have discussed the latter condition. The writer has frequently observed these cecal cores in experimental birds. They interfere with, of course, and often largely prevent cecal discharges, and so make impracticable quantitative studies of cecal coccidiosis on the basis of number of oocysts discharged. As the birds recover from the severe form of the disease, the epithelium in the glands and the tunica propria are restored, but in the most severely afflicted birds the surface epithelium between the glands is not renewed. Thus may be explained some of the permanent adverse effects of the disease.

**Seasonal incidence.** Under ordinary farm conditions, most outbreaks of coccidiosis occur during the months of May, June, July, and August. This is clearly shown by Durant and McDougle (1939) in a graph of 838 autopsied chickens in Missouri. In broiler raising, however, chicks are reared throughout the year, and outbreaks may occur at any time.

**Age factor.** Since cecal coccidiosis is the most widespread of the troublesome ones, most of the investigations have been concerned with it. It occurs principally in young chicks, but seldom in those less than 10 or 11 days old. Many of the worst outbreaks occur at the age of six to eight weeks. Herrick, Ott, and Holmes (1936a) made a study of experimental infections in which they secured many important data concerning the effect of age upon mortality and erythrocyte counts. They found that, in general, the red count was

normal until almost the fifth day of the infection, thence it declined precipitously until the seventh day, after which it climbed steadily until by the twenty-seventh day it again approximated the normal. The heaviest mortality (72 per cent) occurred in chicks one month old, as well as the greatest decrease in erythrocytes (60 per cent decrease). Mortality and red cell decrease were also heavy at the ages of one-half month and two months, while in older birds (three, four, seven, ten, and fifteen months) mortality was low or lacking, though the drop in red cell counts amounted to from 29 per cent to 46.8 per cent. Levine (1940a), in a study of subclinical coccidial infection in pullets at least eight months old, reported the presence of coccidia as follows: *E. mitis, E. acervulina,* or both, 53 per cent; *E. praecox,* 33 per cent; *E. maxima,* 28 per cent; *E. necatrix,* 38 per cent; *E. tenella,* 23 per cent. Thus, the older birds were serving as abundant sources of coccidial infection, although only 8 per cent showed gross lesions of coccidiosis.

**Effect on development and egg production.** As to the effect of coccidiosis upon the future development of birds, Mayhew (1932a, b; 1934b) has found that birds inoculated during the seventh or thirteenth and fourteenth weeks are definitely handicapped in that they do not regain the weight lost during an attack in the following three months; i.e., as compared with the uninfected controls. In a later study he showed that hens developed from chicks inoculated at the age of six to eight weeks laid 19.25 per cent fewer eggs than the controls, and did not attain normal weight (as determined by controls) until five or six months after the attack. Furthermore, severely affected birds begin to lay from six to seven weeks later than the controls. Unfortunately, the species of coccidia concerned in these studies were not stated, but it is apparent that coccidial infections in young birds may leave strikingly unfavorable impressions on the flock for many weeks. It is questionable whether pullets that have had a severe attack of coccidiosis are worth saving. Indeed, Mayhew definitely states that they are not, and there is very much to be said for this opinion.

**Physiological effects.** Severe cecal coccidiosis, with loss of blood, causes a rise in blood sugar during the fifth, sixth, and seventh days of the infection. Artificial bleeding produces the same effect, while starvation does not (Pratt, 1940). Pratt (1941) later found that on the sixth day of the infection the glycogen content of the bird's muscle was less than half of that in normal birds starved 19 hours, while the liver glycogen was slightly higher, though more variable than normal.

Waxler (1941a) has demonstrated that feeding concentrated physiological salt solution to birds during the hemorrhagic phase of the disease effects a lesser rise in blood sugar than when no salt is fed. In addition, the mortality rate was three times greater in the untreated birds than in the salt-fed. He could not, however, positively attribute the beneficial effects of salt feeding to the lower rise in blood sugar.

**Prevention.** The ideal control of the poultry coccidioses would seem to consist simply of preventing the ingestion of viable sporulated oocysts by susceptible hosts. The difficulties lie in perfecting methods of achieving this desideratum.

The most widely advocated and, where attempted, successful of all are practices having to do with management of brooder and laying houses, yards, and runs. The previously discussed findings of Andrews and Tsuchiya showed that on a poultry farm the greatest concentrations of oocysts are, as one might suspect, built up in places where the birds spend the greatest part of their time; namely, under the roosts or shelters and in the vicinity of apparatus for dispensing food and drink. Devices to prevent contact of the birds with their droppings in such areas should, and in actual practice do, reduce losses from coccidiosis, as Van Es and Olney (1940) indicated when they wrote as follows: "The more or less complete separation of the chicks and their food and water from fecal contacts, by the measures described, is capable of reducing both mortality and coccidial parasitism to a bearable minimum."

Actually, effective practices are sometimes difficult to put in force, and there may be pitfalls, as Van Es and Olney learned. Wire netting stretched horizontally between the roosts and the platform underneath protect the flock from the roost accumulations. Waterers may be mounted on inch hardware cloth. Feeders, likewise, can be mounted over heavy wire netting or hardware cloth. The protected droppings should be removed frequently. The frequent removal and replacement of the litter is recommended also, but despite all these precautions, the problem often remains unsolved. Mechanical convection of the infection by insects may be responsible for some of the failures, but there may frequently be other reasons. Van Es and Olney found that a coarse gravel floor in the lots was not particularly effective in control, but that a hardware cloth floor above the gravel did help materially. Another source of infection in their lots proved to be droppings originating from birds roosting on the tops of feeders of a certain type. The latter difficulty was solved by constructing a new type of feeder that was placed flush with the fence enclosing the lot, with the hopper outside (Van Es and Olney, 1937). The two advantages of this type of feeder were that it eliminated roosting on the top and permitted food to be supplied from the outside without the attendant entering the lot.

The thorough cleansing of the floor and equipment in a brooder house between each group of chicks brooded is helpful. After the thorough removal of filth, floor and utensils should be thoroughly scrubbed with hot lye water. Sawyer and Hamilton (1935) recommended 1 pound of lye to 20 gallons of water. Afterwards floor and equipment should be thoroughly dried for, as Pérard (1925) has demonstrated, drying is probably one of the most potent natural forces in the destruction of oocysts. The blowlamp does

not appear to be a very efficient means of oocyst destruction (Horton-Smith and Taylor, 1939).

Of all the fumigation methods that have been proposed, ammonia fumigation seems the most practicable. Horton-Smith, Taylor, and Turtle (1940) obtained complete killing of the oocysts of *E. tenella* with a 1.0 per cent solution of 0.88 per cent ammonia in 24 hours, with a 5.0 per cent solution in 2 hours, and with a 10 per cent solution in 45 minutes. They describe a method of fumigation with gaseous ammonia.

It is important that the attendant takes care that he does not track fecal material from one house or pen to another. Since thorough drying is so fatal to oocysts, much less infection would probably be disseminated if hands and shoes were kept as clean and dry as possible.

The form of range management advocated is the three-year rotation plan, wherein the range is used for chicks but one year, and planted to a cereal crop and grass the next two years. Under this plan the brooder houses are of the movable type. Each house is thoroughly cleaned before it is transported into the clean range. If range shelters are used they should be floored with 1½-inch mesh wire at least 6 inches from the ground, and when shelters are moved the droppings underneath should be removed.

The "deep litter method" of controlling poultry coccidiosis has recently been tested by Boughton (1939). It consists of the use of a deep layer of sawdust or shavings, stirred daily, over the entire period of brooding, i.e., ten or twelve weeks. The litter is renewed just before the advent of the new brood of chicks. Boughton's preliminary study did show that such litter, when dry, does reduce the potential number of sporulated oocysts through drying. Heavily contaminated litter not artificially moistened served as an excellent sporulating medium for the oocysts, while similar litters kept wet did not serve so well as a medium for sporulation. It was concluded that rapid drying and extreme wetness were unfavorable to sporulation, while a limited amount of moisture was highly favorable. Deep litter undoubtedly brings about a mechanical dilution of the concentrations of oocysts as they are passed, thus reducing in general the size of the infective dose and consequently the severity of the ensuing infection.

More recent improvements in the deep and built-up litter methods consist of better insulation and ventilation of the poultry house, which favor dryness of the litter, and the use of hydrated lime in the litter. Kennard and Chamberlin (1947) have adopted the following procedure for combining the built-up litter and hydrated lime practices in the brooder houses: The floor litter may consist of shavings, straw, ground corn cobs, cut or shredded corn stover, peat moss, or cane. At intervals of two to four weeks hydrated lime is scattered over the litter at the rate of 10 to 15 lb. per 100 square feet of floor space, and carefully mixed with the litter at time of distribution in order to

avoid caustic effects on the feet of the birds. The litter is stirred and re-distributed every 2 or 3 days during the first eight weeks and daily after that. Litter and lime may be added at any time as needed, but lime is seldom needed after the first four or five weeks. Such litter will remain dry and fresh for eight to sixteen weeks. Particular care should be taken to redistribute the litter about the water fountains and feed boxes. A modification of this method is used for the laying house, whose litter need be replaced only once a year. How does the procedure affect the rate of coccidiosis? The Ohio Station had no coccidiosis in a brooder house in a season during which five successive broods totaling 10,000 chicks occupied the structure. Coccidiosis was not infrequent previous to the utilization of hydrated lime.

Of the many attempts that have been made to control (and treat) coccidiosis through supplements to the ration, the use of milk and milk products has received the most attention. Mayhew (1934a) has pointed out that there is considerable confusion as to the terms treatment and prevention in the discussions relating to the feeding of milk or milk products, although it is generally to be understood that treatment is to be started when coccidiosis appears in the flock. Thus, one is left with the impression that the goal in most cases has been flock treatment to save the birds not fatally stricken or not yet diseased. Flock treatment actually amounts to prophylaxis to a considerable degree, though it has also been represented that the condition of birds not too seriously stricken would improve under milk treatment.

So far as the author can ascertain, Fantham (1915) and Beach (1917) were the first to propound the merits of sour skimmed milk and buttermilk in controlling avian coccidiosis. Heavy feeding of dried milk, dried skim-milk, dried buttermilk, lactose, or *Bacillus acidophilus* as a component of the mixed ration has been advocated more prominently, particularly for *E. tenella* infections (Beach, 1925; Beach and Corl, 1925; Beach and Davis, 1925). The following formula (parts by weight) was recommended by Beach and Freeborn (1933) : dry skimmilk or buttermilk, 40; wheat bran, 10; yellow cornmeal, 30; ground barley, 20. This mash is to be offered to the entire flock when bloody droppings are first noted, and no other food stuffs except an amount of grain equal to one-third the weight of mash consumed. Fresh water is to be constantly available. The outbreak is supposed to be checked after this regimen has been practiced for two weeks, when the usual ration is resumed. The merit of this flock treatment has been ascribed to various factors: (1) the creation of an acid condition in the ceca, to which the parasite reacts unfavorably; (2) the growth-stimulating properties of the milk products which confer on the birds sufficient vigor to overcome the infection; (3) the laxative effect of the milks which results in the flushing of great numbers of the parasites from the ceca, thus reducing their depredations; (4) a high-protein environment for some reason or other unsuitable

for the development of the coccidium in the same way that a high-protein ration has been shown to be unfavorable to a number of other species of intestinal protozoa (Hegner, 1923, 1924; Hegner and Andrews, 1925).

The plan of feeding a ration mixture of about 40 per cent dry skimmilk or dry buttermilk has been used with varying results. Johnson (1923) early showed that a diet consisting solely of sour milk did not prevent severe infection and death, and later (1927) he states: ". . . a ration containing forty per cent dried milk does not prevent serious coccidial infection in experimental fowls." He does add, however, that such a ration for a period of one or two weeks is worthy of consideration in flocks showing a severe acute outbreak. Chandler (1932) observed rather high mortality in pens on low powdered buttermilk and high powdered buttermilk rations, but obtained definitely less mortality in experimental infections in chicks on the rations with 15 per cent or more powdered buttermilk. Tyzzer (1929), who fed 20 per cent lactose in the ration, reported that the coccidial infection was in no way checked by the procedure. Neither did feeding a mash wet with skimmilk, together with a constant supply of skimmilk to drink. Moore (1928) seemed to have obtained definite value from feeding 30 per cent powdered buttermilk in the mash. Mayhew (1934a), who started the 40 per cent dried buttermilk mash when the chickens became sick, obtained no beneficial effects on recovery, as measured by weight gains, from the special feeding.

Becker and Wilcke (1938) carried out an experiment with four lots of chicks to ascertain whether the milk treatment had prophylactic value in *E. tenella* infection. Lot 1 received a dry mash of the "ordinary" type, but lacking dry milk and containing in its place 10 parts soybean oil meal; lot 2, a similar mash but with 10 parts dried buttermilk and only 2 parts of the soybean oil meal; lot 3, a ration of the "ordinary" type made up to 40 per cent with dried buttermilk; lot 4, a ration similar to Beach and Freeborn's (see above) except that ground oats replaced the ground barley and 1 part of cod liver oil was added. The chicks commenced to eat the special rations in the sixteenth day of their lives, and 10 days later all were inoculated with *E. tenella.* The least losses took place in lot 4; lot 1 suffered but little worse; losses in group 3 were heavy, but lot 2 suffered the most. A repetition of this experiment (unpublished) gave comparable results except that there were considerably fewer deaths in lot 3 than in lot 4. The experiment showed conclusively that the modified Beach and Freeborn formula did have a distinct prophylactic value in *E. tenella* infection. Curiously, in both experiments there were significantly fewer losses in the lots that received the "ordinary" formula lacking in dried milks but with soybean protein than in the lots that received the dried buttermilk and much less soybean oil meal. In the first experiment 40 parts dry buttermilk in the ordinary ration gave but little evidence of protection, but in the second experiment it gave stronger evi-

dence of such protection, though not so much as ration 4. Thus it appears that it is better to mix the heavy amounts of dried milk in simple formula, such as the modified Beach and Freeborn's, than in the more complicated ones, though the latter may have some degree of value.

Becker and Waters (1938, 1939) made other tests with dried buttermilk and dried skimmilk in the ration of chicks experimentally infected with *E. tenella* some time after they had commenced to subsist on the test rations. In every case there were heavier losses among chicks on the milk mixtures. The practice of feeding mashes lacking milk was not recommended, however, because the birds do not grow so well, and there is danger of complications such as "curly-toe paralysis." Furthermore, a number of experiments by the writer subsequent to the published ones have shown that by no means are uniform results to be expected. Indications are that there is considerable variability either in the properties of the feeding stuffs or in the predisposition of the chicks.

The feeding of sulfur in the ration has been advanced recently as a practice having promising possibilities for the control of *Eimeria tenella* infection.

Herrick and Holmes (1936) carried out two sets of experiments, one with sulfur fed from hatching until infection at the age of three weeks, another with sulfur fed for 6 days prior to infection. In the first case the ingredient was mixed in the ration at the 2, 5, and 10 per cent levels; in the second, at the 10 per cent and 20 per cent levels. Since mortality from experimental *E. tenella* infection was strikingly reduced in both tests, the feeding of sulfur seemed to offer considerable promise. The group of chicks fed sulfur from hatching, however, developed irritation of the cloaca, and their growth rate was markedly retarded. The group fed sulfur for only 6 days prior to infection did not show these adverse effects. Herrick and Holmes later reported that continued feeding of sulfur at the 5 per cent or higher level resulted in rickets when cod liver oil was used as the source of vitamin D. Holmes, Deobald, and Herrick (1938) followed up with a number of comprehensive tests, concluding: "If chicks are fed 2 per cent or more of granular sulfur, wettable sulfur, or flowers of sulfur, and are entirely dependent upon cod liver oil of the quality tested in these trials, rickets is quite likely to develop." Probably the correct estimation of the status of sulfur is that it has merit in coccidiosis control, but ill effects otherwise that have accompanied its feeding to date do not warrant its recommendation in poultry-raising practice. Goff (1943), however, commences sulfur and charcoal feeding when the chicks are four weeks old and continues it until they are twelve to fourteen weeks old. He claims that in this way he avoids the retardation of growth occurring when sulfur is fed to younger chicks.

The relation of the amount of calcium carbonate in the ration to coc-

cidiosis also has a bearing on control. Holmes, Herrick, and Ott (1937) showed quite clearly that when this ingredient was fed at the 6 per cent level, mortality was considerably higher than when it was either not fed or held at the 3 per cent level. The feeding of calcium carbonate to growing chicks at high levels is a questionable practice, since Halpin has shown that 2 per cent is sufficient in the usual chick ration.

Herrick, Holmes, and Degiusti (1942) have continued their search for forms of sulfur or organic sulfur compounds that might prove more success-ful than the sulfur they tested. Pertinent details of the work are too numerous to be presented here, but of ten synthetic nitrogen-sulfur compounds tested, three gave protection against cecal coccidiosis. The most satisfactory of all was tetraethylthiuram monosulfide. It gave complete protection against cecal coccidiosis when a single dose of .0004 cc. (but Table V shows .0003 cc.) per gram of body weight was administered 6 hours before the birds were infected with oocysts. Results were not satisfactory when this and the other drugs were fed continuously.

**Treatment.** Milk treatment is also held by some to be of value in treating sick birds as individuals, providing they are not beyond hope of recovery. Johnson (various publications) thinks it has a certain value in cecal coccidio-sis, while Tyzzer (1937) admits the possibility of certain benefits in *E. prae-cox* infection. Mayhew (1934a) found no curative value in powdered butter-milk. If there is any therapeutic value in feeding milk preparations, it is probably not due to their rendering the intestinal content more acid than normal for, according to Kerr and Common (1935), the latter does not occur. Sulfur treatment appears to be definitely excluded, for Herrick and Holmes (1936) found it had no value in cecal coccidiosis if started later than 2 days before the date of infection.

There have been many drug treatments suggested for fowl coccidiosis. The following is a list of some of the materials recommended: terpentine mixed with castor oil, potassium dichromate, potassium permanganate, copperas, copper sulfate, mixture of iron sulfate and glycerine, sulfo carbo-lates, methylene blue and calomel, Epsom salt, sodium bicarbonate, bismuth subnitrate, colloidal iodine (Chandler, 1929), mercuric chloride, quinine, creolin, carbon tetrachloride, hydrochloric acid, azamine, tannic acid, alum, powdered ipecac, acetic acid, and vinegar. (See also Krijgsman, 1929c, and Chandler, 1932.) Kerr and Botham (1931) claimed success with iodine in milk, but Grzimek (1931) found it valueless. The difficulty with most of the drug treatments until recently has been that they have little or no experi-mental basis to recommend them, and many of them, when carefully tested, have proved of no value.

The advent of the sulfonamides marks a new era in suppressive and therapeutic drug treatment of coccidiosis. Levine (1939) made the inter-

esting discovery that sulfanilamide suppressed the normal development of
*E. mitis, E. hagani, E. praecox, E. acervulina,* and *E. maxima,* so that oocysts
did not appear at the anticipated date, though a reduced number of the
terminal stages appeared a few days after treatment was discontinued. There
was no effect of this drug upon *E. tenella* and *E. necatrix,* the two most patho-
genic species which, incidentally, produce schizonts deep in the submucosa.
Sulfapyridine's action almost paralleled that of sulfanilamide (Levine,
1940b). Sulfapyridine, unlike sulfanilamide, did not show evidence of
toxicity for the chicken. Three groups of birds were fed 0.7 per cent sulfa-
pyridine in the mash for 3 days prior to the feeding of infective oocysts and
throughout the balance of the experiment with no indications of toxicity.
However, it was pointed out that both drugs were too expensive for general
field use.

The partial success attained with sulfanilamide and sulfapyridine
(neither drug affected *E. tenella or E. necatrix*) encouraged Levine (1941a)
to further tests on poultry coccidia with related drugs. His unpublished data
indicated that sodium-2 sulfanilamidobenzoate monohydrate, sodium di-
sulfanilamide, and sodium sulfanilyl sulfanilate were ineffective. Sulfa-
guanidine, however, when mixed with the ration at the ½ per cent level,
prevented discharge of oocysts from chickens inoculated with *E. praecox,*
*E. mitis, E. maxima,* and *E. hagani.* At the 1 per cent level it markedly re-
duced the severity of symptoms and lesions due to *E. tenella,* while a 1½
per cent concentration was effective against *E. necatrix.* The experiments
were well controlled, and the data prove conclusively the effectiveness of the
drug as a prophylactic, but it had no curative effect on chickens already
infected with *E. tenella* and *E. necatrix.* Later, Levine (1941b), in a sum-
mary of the comparative effects of sulfur, sulfanilimide, sulfapyridine, sulfa-
thiazole, and sulfaguanidine, suggested that, since the sulfa drug mentioned
must be fed before exposure to infection takes place if beneficient results
are to be obtained, sulfaguanidine might be used under circumstances where
it is apparent that exposure of the flock to lethal doses of coccidia is immi-
nent, and that the same drug might also be used to aid birds in acquiring
active immunity to coccidiosis.

Farr and Allen (1942), Horton-Smith (1942), and Allen and Farr (1943)
have attested to the prophylactic value of sulfaguanidine against cecal coc-
cidiosis in mash containing 1 or 2 per cent of the drug, providing the treat-
ment is instituted several days before ingestion of infective oocysts and con-
tinued for more than a week thereafter. It would seem from the work of
Waletzky and Hughes (1946) that 0.75 per cent might be the minimal re-
quirement for marked prophylactic value. The latter work should be read
by all interested in sulfonamide activity in avian coccidiosis.

A new chapter was written in the therapy of coccidiosis when Horton-

Smith and Taylor (1942, 1943, 1945) obtained beneficial results with sulfamezathine (=sulfamethazine) and sulfadiazine in the food or drinking water after establishment of the cecal infection in chicks through induced epizootics. Treatment reduced mortality by 50 to 73 per cent of that in the controls. Hawkins (1943) confirmed this work in general, and noted inhibition of the cecal coccidiosis by substituting a saturated solution of sulfamethazine for drinking water 96 hours after infection. Hawkins and Kline (1945) found that 0.4 to 1.0 per cent in the feed, started 4 days after infection, gave more constant results than solutions in drinking water. Farr and Wehr (1947) and Wehr and Farr (1947) attributed the beneficial effects of sulfamethazine in *E. tenella* infection to the susceptibility of the second generation schizonts to the toxic effects of the drug.

Ripsom and Herrick (1945) found that a single dose of sulfadiazine (0.5 gm.) afforded greatest protection to very young chicks when given on the third day of the *E. tenella* infection, and that 0.1 per cent in the mash commencing immediately before inoculation and continued for 7 days afforded complete protection. They also found no oocysts were produced in the ceca of chicks receiving the mash commencing the fifth day.

Swales (1944, 1946a) confirmed Horton-Smith's work on sulfamezathine (=sulfamethazine), and showed that a related compound, sulfamerazine and its sodium salt, were highly coccidiostatic and would check the disease when treatment was started at the first sign of bloody droppings in the flock. The levels found satisfactory were 2 gm. of sulfamerazine per pound of dry mash or 2 gm. of its soluble sodium salt per liter of drinking water. There was no advantage in continuing the treatment for more than 3 days. (See also Hawkins and Rausch, 1946.) Horton-Smith and Boyland (1946) have effectively treated *E. tenella* infections with 0.2 per cent sodium sulfamezathine or 0.1 per cent sodium sulfapyrazine in the drinking water. Treatment with the former was begun 24, 48, 72, and 96 hours after infection and continued 3 days after the last deaths from coccidiosis. Their tests showed that for curative action in *E. tenella* infection sulfamezathine and sulfapyrazine were superior to sulfadiazine or sulfamerazine, both of which, however, had more therapeutic effect. Asplin, Boyland, and Horton-Smith (1946) have issued the warning, however, that because of the dangers involved in long-continued administration, sulfamezathine treatment should not exceed one week.

The action of sulfonamides in coccidial infection is antagonized by para-aminobenzoic acid (cf. Horton-Smith and Boyland, 1946; Waletzky and Hughes, 1946).

Other forms of drug-prophylaxis or therapy against cecal coccidiosis for which promising claims have been made are the following: (1) borax in the mash (2.0 per cent) or drinking water (0.3 per cent) (Hardcastle and Foster, 1944); (2) certain halogenated arsonic acids and their sodium salts

at the proper concentrations in feed or water (Morehouse, 1946; and Morehouse and Mayfield, 1946) ; (3) triethanolamine hydrochloride at the 5 per cent level (Harwood and Guthrie, 1943) ; and (4) a sulfur-urea mixture at the 2 per cent level in the mash (Harwood and Guthrie, 1943). During the next few years it will have to be decided which drugs are actually effective in the control of poultry coccidiosis and which of them, if any, can safely and economically be used.

## REFERENCES

Allen, E. A.: 1932. The influence of diet on the development of experimental coccidiosis in chickens kept under sanitary conditions. Am. Jour. Hyg. 15:163.

———: 1933. The pathogenicity of *Eimeria mitis* Tyzzer, 1929, to 3-month-old chickens. Jour. Parasit. 20:73.

———: 1934. A case of prolonged cecal coccidiosis. Proc. Helminth. Soc. Wash. 1:66.

Allen, R. W., and Farr, M. M.: 1943. Sulfaguanidine as a prophylactic during the period of acquirement of resistance by chickens to cecal coccidiosis. Am. Jour. Vet. Res. 4:50.

Asplin, F. D., Boyland, E., and Horton-Smith, C.: 1946. Treatment of caecal coccidiosis of chickens by sulphonamides. Biochem. Jour. 40:ii.

Andrews, J. M.: 1927. Host-parasite specificity in the coccidia of mammals. Jour. Parasit. 13:183.

Andrews, J., and Tsuchiya, H.: 1931. The distribution of coccidial oöcysts on a poultry farm in Maryland. Poultry Sci. 10:320.

Balozet, L.: 1933. Sur une coccidie de la mangouste. Bul. Soc. Path. Exot. 26:913.

Barger, E. H., and Card, L. E.: 1935. Diseases and Parasites of Poultry. Lea and Febiger, Philadelphia.

Beach, J. R.: 1917. Bacillary white diarrhoea or fatal septicemia of chicks and coccidiosis or coccidial enteritis of chicks. Calif. Agr. Exper. Sta., Circ. 162.

———: 1925. The effect of feeding *Bacillus acidophilus*, lactose, dry skimmilk, or whole milk on the hydrogen ion concentration of the contents of the caeca of chickens. Hilgardia 1:145.

———: 1932. Coccidiosis of chickens. No. Am. Vet. 13:27.

——— and Corl, J. C.: 1925. Studies in the control of avian coccidiosis. Poultry Sci. 4:83.

——— and Davis, D. E.: 1925. The influence of feeding lactose or dry skimmilk on artificial infection of chicks with *Eimeria avium*. Hilgardia 1:167.

——— and Freeborn, S. B.: 1933. Calif. Agr. Ext. Serv., Circ. 8:47.

Beaudette, F. R.: 1928. The poultry range may be dangerous. N. J. Agr. 10:3.

Becker, E. R.: 1933. Cross-infection experiments with coccidia of rodents and domesticated animals. Jour. Parasit. 19:230.

———: 1934. Coccidia and Coccidiosis of Domesticated, Game, and Laboratory Animals and of Man. The Iowa State College Press, Ames, Ia.

———: 1940. Coccidioses of domesticated birds, with special reference to the common fowl. Vet. Med. 35:401.

——— and Waters, P. C.: 1938. The influence of the ration on mortality from caecal coccidioses in chicks. Ia. State Coll. Jour. Sci. 12:405.

——— and Waters, P. C.: 1939. Dried skim milk and other supplements in the ration during caecal coccidiosis of chicks. Proc. Soc. Exper. Biol. and Med. 40:439.

——— and Wilcke, H. L.: 1938. The influence of dried buttermilk in rations on fatality with coccidiosis in chicks. Poultry Sci. 17:405.

Boughton, D. C.: 1929. A note on coccidiosis in sparrows and poultry. Poultry Sci. 8:184.

———: 1933. Diurnal gametic periodicity in avian Isospora. Am. Jour. Hyg. 18:161.

———: 1937a. Notes on avian coccidiosis. Auk 54:500.

———: 1937b. Studies on oöcyst production in avian coccidiosis. II. Chronic Isosporan infections in the sparrow. Am. Jour. Hyg. 25:203.

———: 1937c. Studies on oocyst production in avian coccidiosis. III. Periodicity in the oocyst production of Eimerian infections in the pigeon. Jour. Parasit. 23:291.

———: 1939. Studies on the control of poultry coccidiosis. I. The sporulation of oöcysts in various types of litter. Bul. Univ. Ga., Vol. 39, No. 8.

Boughton, D. C., Boughton, R. B., and Volk, J.: 1938. Avian hosts of the genus Isospora (Coccidiida). Ohio Jour. Sci. 38:149.

——— and Volk, J. J.: 1938. Avian hosts of Eimerian coccidia. Bird-Banding 9:139.

Chandler, W. L.: 1926. Iodine on the poultry farm. Poultry Sci. 6:31.

———: 1929. Further studies on the value of iodine on the poultry farm. Poultry Sci. 9:40.

———: 1932. On the control of caecal coccidiosis in chickens. Mich. St. Coll. Agr. Exper. Sta., Tech. Bul. 127.

Cole, L. J., and Hadley, P. B.: 1910. Blackhead in turkeys: A study in avian coccidiosis. R. I. Sta., Bul. 141:137.

Corcuff, C.: 1928. Recherches sur la spécificité parasitaire des coccidies. Ann. Parasit. Hum. et Comp. 6:404.

Crooks, K. B. M.: 1934. Cross-infection experiments on parasite-free chicks with intestinal coccidia from the rabbit. Jour. Parasit. 20:277.

Dickinson, E. M.: 1939. The effects of variable dosage of sporulated Eimeria acervulina oöcysts on chickens. Poultry Sci. 18:404.

Dieben, P. A.: 1924. Over de morphologie en biologie van het rattencoccidium Eimeria nieschulzi n. sp., etc. Thesis Veeartsenijkundige Hoogeschool, Utrecht.

Delaplane, J. P., and Stuart, H. O.: 1933. The common house-fly as other than a simple mechanical carrier of avian coccidia. Poultry Sci. 12:390.

——— and Stuart, H. O.: 1935. The survival of avian coccidia in soil. Poultry Sci. 14:67.

Durant, A. J., and McDougle, H. C.: 1939. Coccidiosis in chickens and other birds. Univ. Mo. Agr. Exper. Sta., Bul. 411.

Edgar, S. A., and Herrick, C. A.: 1944. Feeding habits in relation to severity of cecal coccidiosis. Poultry Sci. 23:30.

Ellis, C. C.: 1938. Part I. Studies of the viability of the oöcysts of Eimeria tenella, with particular reference to conditions of incubation. Cornell Vet. 28:267.

Fantham, H. B.: 1915. Coccidiosis in poultry and game birds. Jour. Bd. Agr. London, Vol. 21, No. 10:889.

Farr, M. M.: 1943. Resistance of chickens to cecal coccidiosis. Poultry Sci. 22:277.

——— and Allen, R. W.: 1942. Sulfaguanidine feeding as a control measure for cecal coccidiosis of chickens. Jour. Am. Vet. Med. Assn. 100:47.

——— and Wehr, E. E.: 1947. Developmental stages in the life cycle of Eimeria tenella affected by sulfamethazine treatment. Proc. Helminth. Soc. Wash. 14:2.

Fish, F.: 1931. The effect of physical and chemical agents on the oöcysts of Eimeria tenella. Science 73:292.

Goff, O. E.: 1943. Coccidiosis prevention and control in chickens by the use of sulphur. Timely Poultry Topics (La. St. Univ.), 1943, Vol. 3, No. 1.

Grzimek, B.: 1931. Jodmilch gegen Kückenkokzidiose. Arch. f. Geflügelk. 5:287.

Hadley, P. B.: 1910. Studies on avian coccidiosis. III. Coccidiosis in the English sparrow and other wild birds. Zentralbl. f. Bakt. I Orig. 56:522.

Hardcastle, A. B., and Foster, A. O.: 1944. Notes on a protective action of borax and related compounds in cecal coccidiosis of poultry. Proc. Helminth. Soc. Wash. 11:60.

Harwood, P. D., and Guthrie, J. S.: 1943. Triethanolamine hydrochloride and mixtures of micronized, wettable sulphur with urea for the control of experimental coccidiosis of chickens. Proc. Helminth. Soc. Wash. 10:90.

Hawkins, P. A.: 1943. Sulfamethazine treatment of cecal coccidiosis. Poultry Sci. 22:459.

——— and Kline, E. E.: 1945. The treatment of cecal coccidiosis with sulfamethazine. Poultry Sci. 24:277.

——— and Rausch, R.: 1946. Sodium sulfamerazine in the treatment of cecal coccidiosis. Poultry Sci. 25:184.

Hegner, R. W.: 1923. The effects of changes in diet on the incidence, distribution, and numbers of certain intestinal protozoa of rats. Am. Jour. Hyg. 3:180.

———: 1924. The relations between a carnivorous diet and mammalian infections with intestinal protozoa. Am. Jour. Hyg. 4:393.

——— and Andrews, J. M.: 1925. Effects of a carnivorous diet on the intestinal pH of rats with reference to flagellates. Am. Jour. Hyg. 5:557.

Henry, D. P.: 1931. Species of coccidia in chickens and quail in California. Univ. Calif. Pub. Zool. 36:157.

Herrick, C. A.: 1934. The development of resistance to the protozoan parasite, Eimeria tenella. Jour. Parasit. 20:329.

————: 1935. Resistance of the oöcysts of *Eimeria tenella* to incubator conditions. Poultry Sci. 14:246.

———— and Holmes, C. E.: 1936. Effects of sulphur on coccidiosis in chickens. Vet. Med. 31:390.

————, Holmes, C. E., and Degiusti, D. L.: 1942. The experimental use of organic sulfur compounds for the prevention of cecal coccidiosis in chickens. Am. Jour. Vet. Res. 3:117.

————, Ott, G. L., and Holmes, C. E.: 1936a. Age as a factor in the development of resistance of the chicken to the effects of the protozoan parasite, *Eimeria tenella*. Jour. Parasit. 22:264.

————, Ott, G. L., and Holmes, C. E.: 1936b. The chicken as a carrier of the oöcysts of the coccidia, *Eimeria tenella*. Poultry Sci. 15:322.

Holmes, C. E., Herrick, C. A., and Ott, G. L.: 1937. Studies in coccidiosis in chickens: Calcium carbonate additions and coccidia. Poultry Sci. 16:335.

————, Deobald, H. J., and Herrick, C. A.: 1938. Sulphur and rickets. Poultry Sci. 17:136.

Horton-Smith, C.: 1942. Sulphaguanidine in avian coccidiosis. Vet. Record 54:259.

———— and Boyland, E.: 1946. Sulphonamides in the treatment of caecal coccidiosis of chickens. Brit. Jour. Pharm. and Chem. 1:139.

———— and Taylor, E. L.: 1939. The efficiency of the blow-lamp for the destruction of coccidial oöcysts in poultry-houses. Vet. Record 51:839.

———— and Taylor, E. L.: 1942. Sulphamethazine and sulphadiazine treatment in caecal coccidiosis of chickens. Vet. Record 54:516.

———— and Taylor, E. L.: 1943. Saturated solution of sulphamethazine as a substitute for drinking water in the treatment of caecal coccidiosis in chickens. Vet. Record 55:109.

———— and Taylor, E. L.: 1945. Sulphamezathine in the drinking water as a treatment for caecal coccidiosis in chickens. Vet. Record 57:35.

————, Taylor, E. L., and Turtle, E. E.: 1940. Ammonia fumigation for coccidial disinfection. Vet. Record 52:829.

Jankiewicz, H. A., and Scofield, R. H.: 1934. The immunization of chicks to caecal coccidiosis under conditions of poor sanitation. Los Angeles County Livestock Dept., Los Angeles, Calif.

Johnson, W. T.: 1923. Avian coccidiosis. Poultry Sci. 2:146.

————: 1927. Coccidiosis studies. Third World's Poultry Cong.:330.

————: 1938. Coccidiosis of the chicken with special reference to species. Ore. Agr. Exper. Sta., Bul. 358:1.

Kennard, D. C., and Chamberlin, V. D.: 1947. Lime treatment of floor litter for chickens. Bimonthly Bul., Ohio Agr. Exper. Sta. 32:11.

Kerr, W. R., and Botham, G. H.: 1931. Iodine in the control and treatment of avian coccidiosis. Vet. Jour. 87:10.

———— and Common, R. H.: 1935. The effects of certain acid treatments for coccidiosis on the H ion content of the fowl's intestine. Vet. Jour. 91:309.

Krijgsman, B. J.: 1929a. Übertragung und Prophylaxis der Kokzidiose. Zentralbl. f. Bakt. I, Abt. Orig. 111:438.

————: 1929b. Overbringing en prophylaxis der coccidiose. Tijdschr. Diergeneesk. 56:1032 (Abst. in Biol. Abst., April, 1931).

————: 1929c. Durch Protozoen verursachte Krankheiten. In van Heelsbergen, T.: Handbuch der Geflügelkrankheiten und der Geflügelzucht. Ferdinand Enke, Stuttgart. Pp. 330–73.

Levine, P. P.: 1938. *Eimeria hagani* n. sp. (Protozoa : Eimeriidae) a new coccidium of the chicken. Cornell Vet. 28:263.

————: 1939. The effect of sulfanilamide on the course of experimental avian coccidiosis. Cornell Vet. 29:309.

————: 1940a. Sub-clinical coccidial infection in chickens. Cornell Vet. 30:127.

————: 1940b. The effect of sulfapyridine on experimental avian coccidiosis. Jour. Parasit. 26:233.

————: 1940c. The initiation of avian coccidial infection with merozoites. Jour. Parasit. 26:337.

————: 1941a. The coccidiostatic effect of sulfaguanidine (sulfanilylguanidine). Cornell Vet. 31:107.

————: 1941b. Chemotherapy in the control of avian coccidiosis. Proc. U. S. Livestock San. Assn., 1941.

————: 1942a. The periodicity of oocyst discharge in coccidial infections of chickens. Jour. Parasit. 28:346.

————: 1942b. Excystation of coccidial oocysts of the chicken. Jour. Parasit. 28:426.

————: 1942c. A new coccidium pathogenic for chickens, *Eimeria brunetti* n. sp. (Protozoa: Eimeriidae). Cornell Vet. 32:430.

Mayhew, R. L.: 1932a. Studies on coccidiosis. I. The effects of coccidiosis upon the weights of chickens artificially inoculated during the seventh week. Poultry Sci. 11:34.

———: 1932b. *Ibid.* II. The effects of coccidiosis upon the weights of chickens artificially inoculated during the thirteenth and fourteenth weeks. *Ibid.* 11:102.

———: 1932c. *Ibid.* III. Observations on paralysis with special reference to coccidial infection. *Ibid.* 11:289.

———: 1933. *Ibid.* IV. Mortality and infection among artificially inoculated chickens. *Ibid.* 12:206.

———: 1934a. *Ibid.* V. Treatment with powdered buttermilk. Jour. Parasit. 20:230.

———: 1934b. *Ibid.* VI. Effect of early attack on egg production. Poultry Sci. 13:148.

———: 1934c. *Ibid.* VII. Effects of starvation and removal of caeca. *Ibid.* 13:360.

———: 1934d. *Ibid.* VIII. Immunity or resistance to infection in chickens. Jour. Am. Vet. Med. Assn. 85:729.

———: 1934e. Some practical results of experiments on coccidiosis in chickens. La. Sta., Circ. 7:1.

———: 1937. Studies on coccidiosis. IX. Histopathology of the caecal type in the chicken. Trans. Am. Micr. Soc. 56:431.

Metelkin, A.: 1935. The role of flies in the spread of coccidiosis in animals and man. Med. Parasit. and Parasit. Dis. (Moscow) 4:75–82. Abst. in Trop. Dis. Bul. 32:660.

Moore, J. M.: 1928. The transmission of coccidiosis. Mich. St. Bd. of Agr., Ann. Rep.:184.

Morehouse, N. F.: 1938. The reaction of the immune intestinal epithelium of the rat to reinfection with *Eimeria nieschulzi.* Jour. Parasit. 24:311.

———: 1946. The effect of some halogenated arsonic acids and their sodium salts on *Eimeria tenella* infection in chickens. Jour. Parasit. (Dec. suppl.) 32:8.

——— and Mayfield, O. J.: 1946. The effect of some aryl arsonic acids on experimental coccidiosis infection in chickens. Jour. Parasit. 32:20.

Patterson, F. D.: 1933. Cross infection experiments with coccidia of birds. Cornell Vet. 23:249.

Pérard, C. H.: 1925. Recherches sur les coccidies et les coccidioses du lapin. Ann. de l'Inst. Pasteur 39:505.

———: 1933. Sur le rôle des espèces non sensibles dans la propagation des coccidioses. Bul. Acad. Vétér. de France 86:206.

Pratt, I.: 1937. Excystation of the coccidia, *Eimeria tenella.* Jour. Parasit. 23:426.

———: 1940. The effect of *Eimeria tenella* (coccidia) upon the blood sugar of the chicken. Trans. Am. Micr. Soc. 59:31.

———: 1941. The effect of *Eimeria tenella* upon the glycogen stores of the chicken. Am. Jour. Hyg. 34:54.

Ray, H. N.: 1945. On a new coccidium *Wenyonella gallinae* n. sp. from the gut of the domestic fowl, *Gallus gallus domesticus* Linn. Current Sci. 14:275.

Ripsom, C. A., and Herrick, C. A.: 1945. Effects of various sulfa compounds on the protozoan parasite, *Eimeria tenella.* Jour. Parasit. 31:98.

Roudabush, R. L.: 1935. Merozoite infection in coccidiosis. Jour. Parasit. 21:453.

———: 1937. The endogenous phases of the life cycles of *Eimeria nieschulzi, E. separata,* and *E. miyairii,* coccidian parasites of the rat. Ia. St. Coll. Jour. Sci. 11:135.

Sawyer, C. E., and Hamilton, C. M.: 1935. Coccidiosis in chickens. Poultry Pointers No. 6 (Revised). St. Coll. of Wash. Ext. Serv.

Smith, T., and Smillie, E. W.: 1917. Note on coccidia in sparrows and their assumed relation to blackhead in turkeys. Jour. Exper. Med. 25:415.

Swales, W. E.: 1944. On the chemotherapy of caecal coccidiosis (*Eimeria tenella*) of chickens. Canad. Jour. Res., D, 22:131.

———: 1946a. On the chemotherapy of caecal coccidiosis (*Eimeria tenella*) of chickens. II. Further studies on the use of drugs in established infections. Canad. Jour. Comp. Med. and Vet. Sci. 10:3.

———: 1946b. The chemotherapy of caecal coccidiosis (*Eimeria tenella*) of chickens. IV. Experiments on the use of chemotherapy during the immunizing exposure of chicks. Jour. Am. Vet. Med. Assn. 108:393.

———: 1947a. New methods of controlling caecal coccidiosis in chicks. Canad. Jour. Comp. Med. and Vet. Sci. 11:5.

———: 1947b. Method of controlling caecal coccidiosis of chicks. Publ. 788, Farmer's Bul. 141. Dom. of Canada, Dépt. Agr.

Tyzzer, E. E.: 1929. Coccidiosis in gallinaceous birds. Am. Jour. Hyg. 10:269.

———: 1932. Criteria and methods in the investigation of avian coccidiosis. Science 75:324.

———, Theiler, H., and Jones, E. E.: 1932. Coccidiosis in gallinaceous birds. II. A comparative study of species of Eimeria of the chicken. Am. Jour. Hyg. 15:319.

———: 1937. A discussion of factors influencing the course of coccidiosis. Jour. Am. Vet. Med. Assn. 90:341.

Uhlhorn, E.: 1926. Übertragungsversuche von Kaninchencoccidien auf Hühnerkücken. Arch. Protistenk. 55:101.

Van Es, L., and Olney, J. F.: 1937. The evolution of a sanitary type of chick feeder. Nebr. Agr. Exper. Sta., Bul. 306:1.

——— and Olney, J. F.: 1940. An inquiry into the influence of environment on the incidence of poultry diseases. Ibid., Res. Bul. 118.

Venard, C.: 1933. Helminths and coccidia from Ohio bobwhite. Jour. Parasit. 19:205.

Waletzky, E., and Hughes, C. O.: 1946. The relative activity of sulfanilamides and other compounds in avian coccidiosis (Eimeria tenella). Am. Jour. Vet. Res. 7:365.

Warner, D. E.: 1933. Survival of coccidia of the chicken in soil and on the surface of eggs. Poultry Sci. 12:343.

Waxler, S. H.: 1941a. The effect of feeding concentrated physiological saline to chickens during cecal coccidiosis. Trans. Am. Micr. Soc. 60:453.

———: 1941b. Immunization against cecal coccidiosis in chickens by the use of X-ray attenuated oocysts. Jour. Am. Vet. Med. Assn. 99:481.

Wehr, E. E., and Farr, M. M.: 1947. Effect of sulfamethazine on the coccidian parasite, Eimeria tenella, of chickens. Proc. Helminth. Soc. Wash. 14:1.

West, J. I.: 1940. Coccidiosis of domesticated animals and fowls. Jour. Am. Vet. Med. Assn. 96:603.

Yakimoff, W. L., and Iwanoff-Gobzem, P. S.: 1931. Zur Frage der Infektion der Tiere mit heterogenen Kokzidien. Zentralbl. f. Bakt., I. Orig. 122:319.

———, Iwanoff-Gobzem, P. S., and Buewitsch, B. L.: 1932. Ibid. II. Mitteilung. Zentralbl. f. Bakt., I. Orig. 125:469.

——— and Gousseff, W. F.: 1933. Ibid. III. Mitteilung. Zentralbl. f. Bakt., I. Orig. 129:506.

———, Iwanoff-Gobzem, P. S., and Matschoulsky, S. N.: 1936. Ibid. IV. and V. Mitteilung. Zentralbl. f. Bakt., I. Orig. 137:299.

——— and Matikaschwili, I. L.: 1932. Coccidiosis of skunks. Ann. Trop. Med. and Parasit. 26:539.

## COCCIDIOSIS OF THE TURKEY[1]

Coccidiosis is not a major cause of losses among turkeys, but Hinshaw (1937) attributed to it the deaths of 2.0 per cent of 4,020 poults over a three-year period (see also Becker (1934), pp. 108–9). In the early literature blackhead and coccidiosis were confused, so that losses were ascribed to coccidia when Histomonas was probably the cause.

**Etiology.** The work of Tyzzer (1929) has established that the turkey is ordinarily the host of two species of coccidia, neither of which seems to compare in pathogenicity with *Eimeria tenella* of the chicken, and probably others still undetermined species.

*Eimeria meleagridis* Tyzzer, 1927, has long, ellipsoidal oocysts, with average measurements of $23.79 \times 17.38\mu$. Sporulation requires about 24 hours. There are one or two globular inclusions at one of the poles of the sporulated oocyst. Oocysts appear in the droppings 5 days after the infective feeding.

In adult turkeys it is the ceca which are colonized, but in young poults the lower half of the small intestine and the greater part of the large intestine may also be involved. The schizonts parasitize the surface epithelium rather than the glandular epithelium. No evidence has yet been presented for the pathogenicity of this species.

---

[1] See also coccidiosis in chapter on turkey diseases.

Tyzzer (1929) could not infect chickens, pheasants, or quail with this species.

*Eimeria meleagrimitis* Tyzzer, 1929, is said to agree closely in morphology and distribution in the intestine with *E. mitis* of the chicken. The oocysts, however, are slightly larger and more elongated than those of *E. mitis*, with an average size of 18.0 × 15.25μ. There is a globular inclusion at the narrower end of the sporulated oocyst. Oocysts appear in the droppings 6 days after the infective feeding.

The epithelium of the villi of the small intestine is parasitized throughout by the developing phases, particularly the lower portion. Most of the organisms are found deep in the epithelium below the nucleus, but never in the subepithelial tissues. The schizonts measure 6 to 9.5μ × 5.8 to 7.7μ, and produce about sixteen relatively short and thick merozoites.

Poults of about two weeks are commonly parasitized the heaviest. Tyzzer discusses several cases of severe coccidiosis in individual birds where strong suspicion of pathogenicity on the part of this species might have been warranted, but he did not regard coccidiosis of turkeys as a problem of any considerable importance in New England.

*Eimeria dispersa* of quail origin was transmitted to three turkeys by Tyzzer (1929). The second transfer, however, did not succeed, suggesting an imperfect adaptation to the turkey, but the transfer of the same organism from the turkey back to quail was successful.

### REFERENCES

Becker, E. R.: 1934. Coccidia and Coccidiosis of Domesticated, Game, and Laboratory Animals and of Man. The Iowa State College Press, Ames, Ia.
Hinshaw, W. R.: 1937. Diseases of turkeys. Calif. Agr. Exper. Sta., Bul. 613.
Tyzzer, E. E.: 1929. Coccidiosis in gallinaceous birds. Am. Jour. Hyg. 10:269.

### COCCIDIOSIS OF THE GOOSE

More is known concerning coccidiosis of geese than of ducks, but our information is still meagre, particularly regarding the intestinal forms. The goose alone of domestic poultry is afflicted with renal coccidiosis caused by *Eimeria truncata*. The intestinal species *E. anseris* and *E. nocens* are rarely found, but are pathogenic in severe infections. The other intestinal form, *E. parvula,* is of little clinical significance.

### *Eimeria truncata* Railliet and Lucet, 1890

This parasite attacks geese of from three weeks to three months of age. The developing forms are found in the epithelium of the uriniferous tubules. The disease is very acute, lasting but 2 or 3 days, and is almost always fatal. Usually the mortality in a flock is very high. The clinical signs are extreme weakness and emaciation. At autopsy the kidneys show poorly circumscribed, yellowish-white spots of the size of a pin head. The tubular epithelium is extensively destroyed, and the tubules may be filled with oocysts and urates.

The oocysts are truncate at the anterior end, and measure on an average about 18 by 13μ, although larger and smaller forms have been described. At the completion of sporulation, which requires 5 days, a residual body is found lying among the sporocysts.

· This disease was known only in Europe until McNutt (1929) reported an outbreak in Iowa. So far as is known, the parasite is specific to geese.

*Eimeria anseris* Kotlán, 1932

*E. nocens* Kotlán, 1933

*E. parvula* Kotlán, 1933

These three intestine-inhabiting species are known only by the characters of their oocysts. *E. anseris* is pear-shaped, with a micropyle, and measures 16 to 23μ by 13 to 18μ. *E. nocens* has large oocysts, measuring 25 to 33μ by 17 to 24μ. *E. parvula* oocysts are roundish, without micropyles, and measure 10 to 15μ by 10 to 14μ.

## REFERENCES

Kotlán, A.: 1933. Zur Kenntnis der Kokzidiose des Wassergeflugels. Die Kokzidiose der Hausgans. Zentralbl. f. Bakt., I. Orig. 129:11.
McNutt, S. H.: 1929. Renal coccidiosis of geese. Jour. Am. Vet. Med. Assn. 75:365.

## COCCIDIOSIS OF DUCKS

Literature on coccidiosis of ducks is so meagre that it is still impossible to write very much authoritatively on the subject (see Becker, 1934, p. 42). There have been a number of accounts of losses attributed to coccidia, but the etiology of these cases has not always been sufficiently elucidated. It is no longer held that *Eimeria tenella* (or "*E. avium*") of chickens is the cause of the duck disease, for cross-infection experiments involving ducks and other barnyard fowl on the one hand and coccidia having their origin in the common fowl on the other have resulted negatively (Johnson, 1923; Tiboldy, 1933).

Tiboldy (1933) describes oocysts of the genus Eimeria that were oval, elongate oval, or occasionally rounded, possessed a sturdy wall, and measured from 10.8 to 25.0μ by 8 to 12.6μ. It appears that a number of species are involved, but the question of specificity has not been cleared up.

The only described species from ducks is *Tyzzeria perniciosa* Allen, 1936, from the small intestine of its host. It was described from a six-week-old Pekin duck (*Anas domesticus*) obtained from Rinebeck, Long Island, U. S. A. The oocysts of this genus are peculiar in that they are asporocystic (without sporocysts), and after development for 24 hours outside the body they contain eight sporozoites and a large residual mass. The content of the freshly passed oocyst, however, consists of coarsely granular protoplasm completely filling the oocyst. The asexual developing stages appear in the mucosa of the small intestine from the gizzard to the ceca. There are three morphologically distinguishable generations of schizonts after which (about 48 hours

after inoculation) gametocytes make their appearance. Oocysts were first observed in the tissue by the end of the fifth day and in the droppings on the sixth day.

The species is extremely pathogenic, for Allen lost seven of ten very young ducks which she experimentally infected. The symptoms reported are "loss of appetite and weight, weakness manifested by the inability of the bird to stand for any length of time, and continuous crying as if in distress. The last symptom was especially noticeable in baby ducks."

Macroscopic lesions consisted of inflammatory and hemorrhagic areas throughout the small intestine, especially in the upper half. The intestinal wall was thickened and exhibited rounded white spots on the exterior. In severe cases blood and cheesy exudate, but no core, filled the small intestine.

Tissue sections revealed the penetration of the coccidium into the mucosa and submucosa as far as the muscular layers, and extensive tissue destruction.

### REFERENCES

Allen, E. A.: 1936. *Tyzzeria perniciosa* gen. et sp. nov., a coccidium from the small intestine of the Pekin duck, *Anas domesticus* L. Arch. f. Protistenk. 87:262.
Becker, E. R.: 1934. Coccidia and Coccidiosis of Domesticated, Game, and Laboratory Animals and of Man. The Iowa State College Press, Ames, Ia.
Johnson, W. T.: 1923. Avian coccidiosis. Poultry Sci. 2:146.
Tiboldy, B.: 1933. Experimentelle Untersuchungen über die Spezifität der Kokzidien des Hausgeflügels. Extract of thesis, Budapest.

### ENTEROHEPATITIS (BLACKHEAD) [1]

Some phases of blackhead are dealt with in the chapter on diseases of the turkey. This section is concerned mostly with the etiological agent, *Histomonas meleagridis,* and other organisms sometimes accredited with a share in blackhead or clinically similar infections. Just before 1895 it was believed by at least one worker that blackhead of turkeys was a form of "cholera," or "liver trouble," which might be overcome by a more careful selection of gobblers for breeding (see Cushman, 1893). In 1895, however, Theobald Smith seemed to have elucidated the etiology of the disease by the discovery of an amoeba-like microorganism in affected tissues, but within the last few years it seems that there are still many unsolved problems, as will become evident in this discussion.

### *Histomonas meleagridis* (Smith, 1895) Tyzzer, 1920

Smith's (1895) original investigations in Rhode Island pointed to *"Amoeba meleagridis,"* as the cause of blackhead. He described from sections of lesions spherical or slightly oval bodies from 6 to 10µ in diameter, with a small spherical nucleus 2µ in diameter, in some cases with a nucleolus. In the original paper Smith did state that movements characterized as amoeboid had not yet been demonstrated, but in 1910 Smith again studied the fresh microorganisms from the liver (which were from 8 to 15µ in diameter) in a warm chamber and noted slight changes of form (Fig. 35.5). He states:

---

[1] See also Enterohepatitis in Chapter 40.

"Some of the freed parasites . . . pushed out small finger-like pseudopodia, usually one at a time." The arrangement of the organisms singly or in groups in the tissues and unenclosed by a common membrane suggested that multiplication took place as a simple process of division and not as endogenous segmentation. Multiple agamic division, however, was suggested in 1915. The parasites were located in the interstices and lymph spaces of the tissues, but not within the cells.



Fig. 35.5. Liver from turkey affected with blackhead showing the characteristic parasites. ×920. (Biester, Iowa State College.)

Cole, Hadley, and Kirkpatrick (1910), and before them Cole and Hadley in 1908, and Hadley in 1909, became convinced that blackhead was really due to coccidia, and that "Smith's amoebae were really the schizogenous stages of a coccidium, the encysted stages of which were found in the cecal contents of the great majority of turkeys dying of blackhead." Smith (1895), however, had previously observed coccidia in turkeys and considered their possible relationship to blackhead, for he states: "It is very improbable that these bodies stand in any genetic relation to the true microparasites of the disease." The coccidian hypothesis is now so thoroughly disproved that it is of only historical interest, though a number of bulletins still survive that at least partially accredit it.

Hadley and Amison (1911) and Hadley himself (1916a, b) later became advocates of the theory of the flagellate nature of the infection. They believed that a Trichomonas (sp.?) which ordinarily lives the life of a harmless

resident of the intestine of fowls may assume a new role under conditions that lower the resistance of the host ". . . having experienced its first taste of blood its whole nature is changed; it becomes another animal, raging through the tissues . . ." The flagellate was supposed to be pleomorphic, and transition stages to the forms found in lesions of the disease and cysts were purported to have been observed. Tyzzer (1919) has pointed out that Hadley's intermediate forms do not resemble the blackhead organism at all, and his cysts are merely examples of Blastocystis. Hadley also contended that flagellosis (as he called it) of the ceca and liver could not be regarded as an infectious disease, since Trichomonas existed in the intestinal tract as a facultative parasite, and its disease-producing powers were wholly extrinsic to its own physiological organization.

Jowett (1911) in South Africa likewise held the view that a trichomonad, *Trichomonas eberthi* Kent, was a normal inhabitant of the ceca of healthy birds, and that under certain conditions this flagellate became pathogenic and produced blackhead. Hence, the disease to him also was not infectious. As Tyzzer pointed out, Jowett did cautiously advise that sick birds be isolated or killed, that birds dead from the disease be properly disposed of, and that quarters occupied be treated with quicklime, etc.

The presence of flagellates in the cecal content of turkeys had already been noted by Smith in his first paper. It does not appear that he meant to give them more than passing notice, for he refers to them in a footnote as "flagellates in the ceca of healthy turkeys." The work of Tyzzer (1919, 1920a) and others would seem to have completely discredited the trichomonad hypothesis, but, as we shall see later, it has been revived in a new form. It would seem safe to state that Smith's organism stands today as an almost universally accepted etiologic agent in blackhead of turkeys, if not the only etiologic agent.

Tyzzer (1919) first noted an "extra-nuclear body" lying near the nucleus of the microparasite. Radiating from this body and passing over the surface of the nuclear membrane were delicate and fine filament-like rays (or axonemes). Division of the nucleus was observed to be initiated by the division of the extranuclear body into two portions connected by a deeply staining line or paradesmose. The two daughter extranuclear bodies behave as division centers at opposite poles of the nucleus. Nuclear division proceeds to completion, and the paradesmose disappears from view. The division center associated with each new nucleus comes to appear as a double granule. Division of the cytoplasm presumably follows nuclear division.

In his earlier work Tyzzer mentions no flagella arising from the extranuclear body, and he does describe the amoeboid movements of the organism. He had demonstrated its flagellate nature, however, and in 1920 he renamed Smith's organism *Histomonas meleagridis*. In that year he described the

movements of the tissue parasites as observed under the microscope in the warm chamber at 41° to 42° C. These were of two types: (1) pulsating, intra-cytoplasmic movements attributable to the "kinetic apparatus" wholly en-closed in the cytoplasm, and (2) amoeboid movements varying from slight changes of form to the formation of sharp, wavelike pseudopodia. A rudi-mentary flagellum on the surface of the organism was also observed. Tyzzer (1920a), however, states that there is considerable evidence that the parasite migrates through the tissue by amoeboid movement.

Free flagellated forms were first seen by Tyzzer and Fabyan (1922). These were taken from the ceca of experimentally infected turkeys. They state that late in the disease they found in the ceca of turkeys forms of *Histomonas meleagridis* in a considerable proportion of which one or two short flagella were demonstrable. In 1924 Tyzzer found flagellate forms in the ceca of chickens. Drbohlav (1924) obtained the flagellated forms in cultures from the ceca of diseased birds. Flagellate types show a great variety of amoeboid movement and ingest bacteria, cell fragments, and starch grains (Tyzzer, 1924).

Tyzzer (1934) later gave us more light on the nature and behavior of Histomonas. In cecal discharges under optimum conditions it is fairly rounded, but with irregular surface extensions, and exhibits active amoeboid movements and rhythmic rotary movements. At the proper high temperature the rhythmic beat becomes frequent, and a flagellum may be seen beating in one direction toward the body, causing it to rotate counter-clockwise one-fourth to one-third of a full rotation at each stroke. One form was seen with four flagella.

In cultures the organism usually attains a larger size than in cecal dis-charges. At times culture forms are spread out into thin sheets and exhibit active amoeboid movements; at other times they are more rounded and undergo "rhythmic flagellate motility." When brought into contact with surfaces the latter forms may become amoeboid while still exercising their flagella. The flagellated phases may measure from 4.5μ to 25μ in diameter, and flattened amoeboid forms considerably more. The cytoplasm may con-tain starch and bacteria in considerable quantities. While normally uni-flagellate, aflagellate forms are common, and those with two or even four flagella are not infrequently encountered in cultures.

De Volt and Davis (1936) have confirmed in large part Tyzzer's observa-tions on the behavior and nature of the organism from tissues and in cecal discharges and culture.

### TRANSMISSION IN RELATION TO LIFE HISTORY

1. **Direct infection.** From the facts set forth in the preceding paragraph, it is evident that the coccidia and the ordinary flagellates and amoebae of the ceca of fowls have no part in a consideration of the transmission of Histo-

monas-blackhead. A certain degree of pleomorphism of the parasites in the lesions is indicated, however, by the descriptions of Tyzzer (1919, 1920b). Amoeba-like invasion forms with clear blue staining cytoplasm, either with or without ingested inclusions, occur in the early lesions or at the periphery of the older ones where they are engaged in extending themselves between the otherwise normal tissue cells. In the subperipheral area of the lesion are to be found organisms which have apparently lost their motility and entered upon a vegetative existence, and whose cytoplasm is clear and stains but faintly. It is likely that a large proportion of these are degenerating forms.

In the older portions of the lesions are to be found what Tyzzer once called resistant phases, characterized by the possession of a dense surface membrane, and probably also by a loss of water from the cytoplasm. The nucleus appears shrunken and distorted. These resistant forms are eventually enclosed in definite spaces by the reaction of the tissues or taken up in great numbers by giant cells. That these forms are true resistant phases comparable to the cysts of Endamoeba and capable of transmitting the infection seems doubtful from the later work of Tyzzer himself and his collaborators. Tyzzer, Fabyan, and Foot (1921), and Tyzzer and Collier (1925) indicate that the organisms in lesions kept at room temperature will not survive longer than about 24 hours, and that those in the dejecta from infected birds are likewise short-lived, as proved by experimental inoculations. They were also unable to infect three young turkeys by exposure to turkey droppings of the previous season.

Until recently, neither Smith nor Tyzzer had been able to find definite indication of the presence of Histomonas in the cecal contents or discharges of infected birds. Smith (1915) states, "Although the ceca are the chief seats of the disease, and are evidently the region where the tissues are first invaded by way of the digestive tract, a study of the contents has thus far yielded nothing definite." Tyzzer (1919): "The contents of the ceca are remarkably free from the parasite. It has thus far been impossible to identify it in the discharges examined during the life of the infected bird." Tyzzer (1920a) and Tyzzer and Fabyan (1920, p. 236) make similar statements.

These workers, like most of the others, had a feeling that the natural outlets for the parasite were the cecal discharges and the bile, thence the feces. The parasite was supposed to be acquired by the new host in contaminated ingested material. Tyzzer (1919) and Tyzzer and Fabyan (1920) observed that the parasites ("resistant forms") are occasionally found in stained sections in giant cells both on the surface of the cecal mucosa and extending through the epithelium or bile ducts for a short distance along the lumen. It has not yet been proved that the forms found in the giant cells are capable of multiplying in normal turkeys. They may have become irreparably injured by the digestive enzymes of the giant cells. Tyzzer (1924), as

mentioned previously, encountered the flagellated Histomonas in large numbers in the ceca of chicks early and late in the infections, but not during the acute attack. This observation confirms the existence of a free stage in the cecal contents.

Tyzzer's (1919) comment that it is quite probable that elimination of parasites is less marked in the acute stages of the disease than during recovery might tend to explain Smith's (1917) and Smith and Graybill's (1920a) results which indicate that young diseased birds showing active symptoms do not transmit blackhead. They found, however, that young normal turkeys confined with older birds which had passed through an attack were extremely likely to become diseased. Tyzzer (1920a) also exposed a flock of ten 60-day-old turkeys to two infected turkeys for about six weeks, and fed them diseased liver, ceca, and cecal discharges both in the fresh condition and after being kept for 2 days or more. None became infected. These results, along with others, seemed at the time to indicate that the young turkeys can be infected with neither droppings from birds in the acute stage nor diseased tissues.

In a later series of more critical experiments, however, Tyzzer and Collier (1925), succeeded most strikingly in demonstrating that young turkeys could be infected either by feeding them the fresh liver lesions of infected birds or by rectal injections of this material. Furthermore, of a group of eight young normal Heterakis-free turkeys confined with diseased Heterakis-free turkeys, six became infected through the direct ingestion of freshly passed organisms. Tyzzer and Fabyan (1922) had previously produced blackhead lesions, in the absence of Heterakis, in two out of four turkeys by feeding considerable amounts of active liver lesions. De Volt and Davis (1936) prepared an inoculum from infected livers that they used successfully in infecting poults. These latter experiments show that, contrary to the earlier work, direct transmission of blackhead from active cases to normal birds is a proved possibility. Tyzzer in 1934 stated that oral feeding of materials containing Histomonas, such as liver lesions of acute disease or cecal discharges, is somewhat unreliable owing to death of parasites in passage of the alimentary tract.

There have been outbreaks of the disease in both experimental and farm yard flocks which had no contact with other poultry. Smith and Graybill (1920a) showed that incubator-bred turkeys which were kept in outdoor enclosures away from other domesticated birds picked up from the soil coccidia, Heterakis, and other infectious organisms from material which presumably had been deposited by other birds. They also noted that placing young turkeys with an old flock subsequent to removal to new soil resulted in the appearance of blackhead in the new stock, and that soil recently occupied by old turkeys was infectious to young ones. In the absence of any demonstrated cyst stages, these observations were difficult to explain. De Volt and

Davis (1936) have shown most conclusively that infected soil may be the means of transmission.

It should be mentioned here that there is abundant evidence that the blackhead microorganism is not transmitted through the turkey's egg. Curtice (1907) had first made this assertion. Smith (1917) carried along twenty-three incubator-hatched and brooder-raised turkeys for more than six weeks without the appearance of blackhead among them. Since the eggs employed for rearing the birds were from infected flocks, the experiment supplied considerable evidence that the protozoon was not transmitted through the egg. Later Tyzzer and Collier (1925) state that in their experiments they employed sixty-one controls which did not develop blackhead. None of the control birds in Tyzzer's previous experiments contracted the disease. Thus it is evident that Graybill and Smith's (1920) conclusion that their isolated turkeys were picking up the infection from the soil were well founded, since contact with other domesticated fowls and hereditary transmission could be dismissed from consideration. Consequently, they turned their attention to the cecal worm, *Heterakis gallinae*.

**2. The relationship of** *Heterakis gallinae* **to blackhead.** The initial demonstration of the role of Heterakis, the nematode worm commonly inhabiting the ceca of chickens, turkeys, and other fowl was made by Graybill and Smith (1920). First two turkeys were fed feces of adult turkeys and cultures of embryonated Heterakis ova (from chickens) kept in Petri dishes in normal saline solution for 17 days. Both infected turkeys became sick 15 days after ingesting the ova, and both died within a week. Three more turkeys given the same feed contracted blackhead, as did three which received only the embryonated eggs. Negative results were obtained from feeding turkeys the feces only.

It is apparent that Graybill and Smith were of the opinion that the cecal worms, invading the ceca in large numbers, broke down the resistance of the fowl to the protozoan factor which was already present in the ceca, "probably disseminated when the first spontaneous cases occurred in the stock." They suggested that there might likewise be other agents which, when ingested by the turkeys, would prepare the way for invasion of the tissues by the blackhead organism. In the same year, however, Smith and Graybill showed that the disease could be produced in chickens by feeding incubator-hatched chicks an overdose of the eggs of Heterakis which had been placed in 0.5 per cent solution of bichloride of mercury for 30 seconds, washed, and dried. This time they concluded that the evidence pointed to the presence of *Histomonas* (=*Amoeba*) *meleagridis* in the cultures, but they were disinclined to accept this possibility unreservedly and so maintained that the origin of the virus was still undetermined. They actually found amoebae and flagellates in some cultures of worm ova, but the studies of Glaser (1921) later

showed that these were merely free-living protozoa. Tyzzer, Fabyan, and Foot (1921) confirmed in principle the work of Graybill and Smith, and noted likewise that it was not necessary to feed the virus with the embryonated eggs in order to produce the disease.

That the ovum of Heterakis may actually harbor Histomonas inside its shell has received very strong support from the work of Tyzzer and Fabyan (1922). Heterakis material treated for 3 days with 1.5 per cent nitric acid, which rendered the medium bacteriologically sterile, was proved capable of transmitting blackhead when fed to incubator-bred turkeys. Previous and subsequent experiments have shown that the protozoon concerned in blackhead is fragile and nonresistant, incapable of surviving more than 24 hours at room temperature. So the possibility of its having persisted in the acid outside the egg membranes is almost nil. The discharges of blackhead carriers free from embryonated eggs will not transmit the disease after treatment with 1.5 per cent nitric acid (Tyzzer, 1926).

The possibility that the virus is present in the turkey from the time of hatching and incited to tissue invasions by the worms is discredited by three facts: (1) No microscopic study of tissues of young isolated healthy turkeys has demonstrated the parasites; (2) isolated turkeys may naturally have cecal cores associated with inflammation of the cecum or receive injections of melted paraffin into the ceca and still not develop blackhead (Tyzzer, Fabyan, and Foot); and (3) the disease may at times be produced by feeding active lesions or droppings from infected birds with no Heterakis, but it never appears in isolated incubator-bred turkeys receiving sterile feed.

Tyzzer and Fabyan showed that the disease could be produced in turkeys by feeding hen-yard soil which, presumably, contained embryonated Heterakis eggs. The evidence for the culpability of the egg of the cecal worm is thus most convincing. Feeding unembryonated eggs and male Heterakis will not produce the infection. The disease follows the feeding of the embryonated eggs more often when they are pooled from a number of different birds. The weak link in the chain of circumstances incriminating Heterakis eggs is the failure to demonstrate the protozoon inside their membranes. The proof of the existence of a stage of the parasite there would establish the nematode as a true invertebrate intermediate host. Tyzzer (1926) examined large numbers of eggs with negative results, and Glaser (1921), as mentioned above, could not find the Histomonas in infective cultures of the eggs. Tyzzer (1926) has, however, noted invasion by the protozoon of the tissue of a number of half-grown worms from cases of blackhead. At present it is not known whether the acute or chronic form of the disease is most favorable for the acquisition of the infection by the worm.

Failure to find the worms in enormous numbers in the ceca after the ova were fed does not necessarily indicate that the worm had no role in transmis-

sion, for Tyzzer and Fabyan (1922) found evidence that the worms are destroyed in the diseased ceca.

**3. The relationship of the chicken to the dissemination of blackhead among turkeys.** That the chicken may serve as a true biological host for the blackhead parasite has long been known. The earliest reports of such infections in this fowl which the author has been able to find are those of Chester and Robin (1900), Curtice (1907a, 1907b), and Theobald (1907). Curtice states that others besides himself had noted that the common fowl is occasionally subject to blackhead, but he gives no references. He actually observed the causative organism in the tissues of chickens, noted the differences in the course of the disease in the chicken and the turkey, declared that the parasite carried on multiplicative activities in the cecal contents, and concluded from the evidence afforded by his experiments that "the ordinary fowl carry and distribute the amoebae." He recommended that turkeys be kept away from other barnyard fowl as much as possible, and further went oñ record with the declaration that one reason for the popular belief that turkeys cannot successfully be raised in confinement may be that they are often kept with ordinary fowl which give them the parasite—all of which is quite in accord with present-day teaching. Theobald reported the existence of an infectious entero-hepatitis caused by *"Amoeba" meleagridis* among British poultry, but states that the disease had already been observed in Continental Europe. Milks (1908) described enterohepatitis of chickens with the presence of the blackhead parasite in four widely-separated localities in Louisiana. The disease was limited almost entirely to young chicks, and the mortality was as high as from 30 to 50 per cent of those hatched. Since these earlier reports, the infection has been found repeatedly in chickens, so that there is no doubt that it is quite widespread (cf. Higgins, 1915; Tyzzer, 1919, 1924; Tyzzer and Fabyan, 1922; Smith, 1915; Smith and Graybill, 1920a; Kaupp, 1922; Eriksen, 1925). Since the chicken is not only widely raised on farms, but is also commonly infected with the cecal worm, *Heterakis gallinae,* it is to be expected that it would assume considerable importance in the epidemiology of blackhead among turkeys.

The invariably disastrous experiences of Curtice in attempting to raise turkeys with chickens or on soil previously occupied by them have been alluded to above, although Smith (1915) considers that these experiments do not warrant definite conclusions. Smith (1915) himself attempted to raise nine artificially incubated and reared turkey poults near a hen yard. Six of them acquired blackhead, but his later (1917) experiments in which turkeys mingled with chickens did not lend support to any hypothesis pertaining to the órigin of the infection in the common fowl. Tyzzer and Fabyan (1920) infected a turkey by exposure to common fowls. The experiments of Smith

and Graybill (1920b) added nothing definite to the guilt of the chicken. Graybill and Smith (1920) state that the relation of common poultry to outbreaks of blackhead may be accounted for, at least in part, by the fact that they harbor the cecal worm. Although they had been able to produce the disease in turkeys with embryonated Heterakis eggs from chickens without the addition of materials from infected turkeys, they regarded the source of the virus as still problematic. Smith and Graybill (1920b) were able to produce a mild type of blackhead in incubator-bred chicks by feeding disinfected embryonated Heterakis eggs from old hens. They suggested that the egg cultures were the possible source of the protozoon, but considered that problem still unsettled. At any rate, the work of Graybill and Smith and of Smith and Graybill had shown definitely that the common fowl may be culpable in the turkey disease, but whether it might serve as a source of only Heterakis eggs or of both the eggs and the protozoon was still undetermined.

Tyzzer, Fabyan, and Foot (1921) confirmed the experimental production of blackhead by the feeding of Heterakis, and infected a turkey by exposing it to hens. At autopsy 279 worms were found in the cecum, a greater number than was present in any of the experimentally fed turkeys. Tyzzer and Fabyan (1922), as previously mentioned, were able to infect young incubator turkeys by feeding them (1) the disinfected embryonated eggs of Heterakis from the chicken, (2) the liver lesions from a case of blackhead in a chicken, or (3) dirt from hen yards mixed with the food. The experiment indicates that the parasite in the turkey and chicken disease is identical, and that the turkey may acquire both the cecal worm and the protozoan parasite from the chicken.

**4. Transmission through the turkey's egg.** The evidence that the blackhead microorganism is not transmitted through the egg of the turkey has been discussed above. Furthermore, Tyzzer, Fabyan, and Foot (1921) failed in attempts to produce the inoculated disease in the turkey embryo in the egg. (See also Tyzzer, 1934.)

**5. Arthropod carriage.** The possibility of an intermediate host in the transmission of blackhead was considered by Smith in his original report (pp. 24–25). He dismissed the possibility on the basis of his personal observations that not all flocks were infected as they would tend to become if the disease were carried by an insect, for example, and that the disease becomes perpetuated and diffused among neighboring flocks by uninterrupted transmission. The likelihood that the virus was discharged from the sick bird ("perhaps in an encysted form") and ingested with the food and drink of others seemed better to explain the spread of the infection. Curtice (1907b) mentions that a lot of poults which he had raised free from blackhead "undoubtedly ate all sorts of insects." Tyzzer and Fabyan (1920) fed a young

turkey grasshoppers, crickets, and also about 135 "blowflies" (*Calliphora erythrocephala*) which had fed on finely minced lung lesions from a case of inoculated blackhead. The turkey remained normal and continued to grow. Tyzzer, Fabyan, and Foot (1921) confined three turkeys in a cage which served as a very effective fly-trap, catching swarms of blue-bottle flies on which the young turkeys engorged themselves but contracted no disease. They explained the phenomenal growth of these turkeys by the abundance of insect food available. The "blowfly" or "blue-bottle" collects in great numbers to feed on the droppings and discharges of diseased birds and is eagerly devoured by young turkeys. While the evidence that it or any other arthropod plays no direct part in the dissemination of the disease is all negative, it is to be remembered, nevertheless, that the epidemiology of the disease can be fairly well explained without hypothetical insect vectors. The absence of the protozoon in the peripheral circulation would indicate that biting insects, such as certain lice, play no role in transmitting the disease. (See also De Volt and Davis, 1936.)

**6. Other possible factors in the transmission of blackhead.** Smith and Gray-bill (1920a, b) made certain very important observations and experiments on the epidemiology of blackhead among turkeys which seem to require a consideration of additional and perhaps still unknown factors. As an example, an unused horse paddock enclosed by a high iron fence and not occupied by poultry for many years was plowed early one spring and sowed to oats and grass. Blackhead disease and Heterakis appeared among young turkeys kept in this pen. The explanation offered for such outbreaks is the attraction of wild birds in large numbers to the food supply in the turkey enclosures. These birds were supposed to have deposited infective materials which were taken up by the turkeys.

There is no evidence that ordinary wild birds, such as the English sparrow, starling, and robin, can become infected with blackhead. There remains the possibility, but no proof as yet, that sparrows pick up Heterakis eggs with their food in yards where turkeys or hens infected with blackhead are confined and deposit them later with their droppings in an enclosure where healthy turkeys are kept. Eggs thus deposited might be immediately infective if embryonated or become so after development. Even "blowflies" could transport eggs in a similar manner, for this possibility has not been excluded by Tyzzer's experiments on this insect. The earthworm is said by Cram (1927) to be a mechanical vector for the eggs of *Heterakis gallinae*: "Earthworms may ingest the eggs and carry them in the intestine, and birds may become infested by eating such earthworms; the earthworms may also pass these eggs in their casts and thus infect otherwise uninfected ground." There is a possibility that the eggs might be scattered about by the wind during "dust storms." The various methods of dissemination of the viable eggs of

Heterakis here suggested are to be taken merely as hypothetical, much more so the transfer of the disease by such worm eggs. But they do point to problems worthy of serious investigation.

Cram states that Heterakis eggs (from chickens) frozen for 10 days at 0° to 10° F., then kept at this temperature for six months, were found by Riley and James to develop embryos in 75 per cent of those examined; and that, according to Graybill, ova may survive a desiccation period of from 16 to 18 days, and eggs in soil contained live embryos for eight months (cf. Mumford, 1927). Eggs from the host kept under favorable conditions of moisture and temperature may develop to the infective stage in 7 to 12 days.

Another unknown factor in the transmission of blackhead, which is important in control, is the length of time the Heterakis egg harbors the viable microorganism.

**7. Conclusions.** *Histomonas meleagridis* is a common parasite of turkeys and chickens, but is to be found also in certain other birds to be mentioned subsequently. In the chicken at least (Tyzzer, 1924) and possibly also in the turkey (Tyzzer and Fabyan, 1922) the organism may live and multiply as a flagellate in the cecal contents during the period of incubation and for an indefinite period following recovery from the attack. During the acute phase of the disease it penetrates the tissues. Recovered birds may serve as carriers, discharging large numbers of microorganisms daily from the ceca. The organism can be acquired directly by young poults through contamination of the food with the droppings of infected birds, even in the absence of the eggs of the cecal worm (Tyzzer and Collier, 1925). It is not improbable that an acute epidemic in a flock of turkeys may be due to this method of transmission. The eggs of *Heterakis gallinae* eliminated by infected chickens and turkeys, when embryonated, may transmit the infection to healthy birds upon infesting them. The production of the disease with disinfected eggs indicates a resistant stage of the microorganism within the egg membranes. Sporadic outbreaks are probably due to ingestion of the infective worm eggs which have in some manner been disseminated over the soil. The natural means of dispersal of the worm eggs are not definitely understood in all cases, although it is apparent that the common fowl and the turkey are the principal agents.

It should be made clear that the chicken is not an obligatory host of either the protozoon parasite or the cecal worm. Statements to this effect have appeared and are due perhaps to the emphasis which has been placed upon the separation of turkeys and chickens in the control of the disease. The chicken and the turkey occupy similar places in the biological scheme, and it has been abundantly demonstrated that uninfected turkeys can acquire the infection from infected turkeys. On the other hand, it is declared that the common fowl is a carrier of the protozoon and the cecal worm to a greater

extent than is the case with turkeys (Tyzzer, 1927) . For this reason it is more culpable than the turkey itself in the dissemination of blackhead. (See also De Volt and Davis, 1936.)

### CULTIVATION OF THE PARASITE

The flagellate stages from the cecum of chickens recovering from blackhead were cultivated by Drbohlav (1924) over a period of 81 days. Coagulated white of egg slants covered with blood bouillon containing 1 per cent peptone was the medium preferred. The best range of pH was between 7.2 and 7.8. In culture the parasites feed on bacteria. When a blood agar slant is used in place of the white of egg, red cells may be ingested. The identity of the flagellate cultured with the forms found in blackhead was proved by rectal inoculations into chickens, which showed large numbers of the parasites in the cecal discharges within 3 days, and then developed typical blackhead. Tyzzer (1934) , De Volt and Davis (1936) , and Bishop (1938) also obtained very successful cultures of *Histomonas meleagridis* and carried them on for prolonged periods. Bishop's cultures were unique in that they were started from liver lesions. With them she was able to inoculate chicks per os and per anum, producing cecal lesions.

### THE INFECTION IN BIRDS OTHER THAN THE TURKEY

**The chicken.** It was mentioned in the section on transmission that infections in the chicken have been widely reported. (Cf. Chester and Robin, 1900; Curtice, 1907a and 1907b; Theobald, 1907; Milks, 1908; Cole, Hadley, and Kirkpatrick, 1910; Higgins, 1915; Tyzzer, 1919 and 1924; Smith and Graybill, 1920b; Kaupp, 1922; Eriksen, 1925.) The course of the infection in the common fowl usually runs a much milder course than in the turkey, but is otherwise very similar. Smith and Graybill found that in chicks experimentally infected with Heterakis eggs, the initial lesions appeared in the ceca, usually followed by only microscopic focal collections of lymphocytes or yellowish necrotic specks in the liver. The inflammation and thickening of the cecal wall and the subsequent formation of a core resemble somewhat the condition in the turkey disease, but the invasion of the liver lesions bears no comparison in the two birds. Smith and Graybill state that all their chicks would probably have survived had they not been killed, for the processes of repair had been initiated, whereas experimentally infected turkeys usually died.

Reports such as those by Milks, Kaupp, and Eriksen indicate, however, that at times the disease of chickens may run a much more severe course. Kaupp observed the death of forty-two out of forty-three Silver Spangled Hamburg chicks about five weeks of age. Eriksen autopsied a total of twenty-five birds from seventeen Missouri flocks. The losses ranged from one bird in a flock of 350 to more than 50 per cent in two other flocks. At autopsy

cecal and liver lesions were observed, the latter organ occasionally enlarged to several times its normal size and studded with gray or grayish-yellow areas 3 to 8 mm. in diameter which penetrated deeply into the liver tissue. Histomonas was observed in sections of both ceca and liver. The age of the chicks attacked was from seven to ten weeks (cf. observations of Milks). Thus the infection in chickens may assume a serious nature at times, although under ordinary circumstances it appears to be well tolerated by the host.

Tyzzer and Fabyan (1920) found that subcutaneously inoculated infections in chicks were but local and transient, the birds soon recovering. One chick developed a secondary lung lesion.

**The pigeon.** This bird, whose ceca are but small lateral diverticula, is not known to harbor Histomonas naturally. Tyzzer and Fabyan (1920) produced transient but well-defined localized lesions in the subcutaneous tissue and breast muscles by experimental inoculation of organisms from the livers of active cases in turkeys.

**The ruffed grouse.** According to Tyzzer and Fabyan (1920), this bird commonly succumbs to the disease in captivity (cf. also Graybill, 1925).

**The common quail.** According to Tyzzer and Fabyan (1920), this bird is somewhat susceptible. Graybill (1925) states that the disease occurs occasionally in quail.

**The duck.** Tyzzer, Fabyan, and Foot (1921) were unable to produce inoculated blackhead in two Indian runner ducks.

**The English sparrow.** Cole, Hadley, and Kirkpatrick (1910) had placed considerable emphasis upon the sparrow as a disseminator of blackhead. They were laboring under the miscomprehension that blackhead was a form of coccidiosis which was prevalent also among sparrows. Smith and Smillie (1917), however, showed that the sparrow coccidia belong to the genus Isospora, while those found in chickens and turkeys are Eimeria. Tyzzer, Fabyan, and Foot (1921) were unable to produce blackhead lesions in many experimentally inoculated English sparrows.

**The guinea fowl.** Tyzzer, Fabyan, and Foot produced local and self-limited lesions in three guinea chicks by subcutaneous inoculation. Graybill (1925) states that the disease occurs occasionally in this bird.

**The pheasant.** Tyzzer, Fabyan, and Foot produced a small, local, persistent lesion in a day-old pheasant. When killed no other lesions were noted. Graybill (1925) states that the disease occurs occasionally in this bird. In a personal communication, Tyzzer states that in connection with the New England ruffed grouse investigation, he has attempted to ascertain whether the ring-necked pheasant was an agent in the spread of blackhead. Pheasants show great numbers of Heterakis, but he has never been able to produce blackhead in turkeys by feeding the Heterakis eggs from pheasants. Neither has he been able to produce carriers by the rectal inoculation of half grown

pheasants with blackhead. Cecal material collected from pheasants and inoculated per rectum into young turkeys has always failed to infect the latter.

**The peafowl.** Graybill (1925) states that the disease occurs occasionally in this bird. Dickinson (1930) examined post-mortem two peafowls which had presumably become infected from running with a flock of turkeys. Both showed the liver and cecal lesions typical of fatal blackhead.

**Mammals.** According to Tyzzer and Fabyan (1920), rabbits, guinea pigs, mice, and Japanese waltzing mice are not susceptible to the inoculated organism. It is likely that all mammals are resistant.

### "Blastocystis"

A few years ago Enigk (1935) in Germany made the astonishing announcement that Histomonas was not involved in the etiology of fifty-six cases of blackhead in young turkeys and of five in young chickens, but that in these cases the disorder was attributable instead to a budding fungus of the type commonly designated "Blastocystis," a morphological description of which is given by him. He succeeded in cultivating this Blastomycete from the tissues of thirty-six of the poults, thirty-three of which were from the liver. His cultures were capable of producing blackhead when inoculated into normal birds. The culture medium was maltose-agar.

He found the fungus to occur frequently in the alimentary tract of sound birds, as well as in birds which had died of other causes. He speculated that perhaps injury to the cecal mucosa, as through the activities of the nematode Heterakis, might permit penetration of the fungus into the tissues, and that lowering of the host's resistance through some other cause might be the determining factor in producing the disease. Menzani (1933) had also considered a Blastocystis to be the primary invader. Enigk's hypothesis of blackhead produced through the agency of a Blastomycete has a certain plausibility. A notable uncertainty has always existed concerning the nature of the parasitic bodies in blackhead liver, as Enigk explains. Smith's (1915) perplexity concerning their amoeboid movements, the variability of the parasite in size and appearance (Tyzzer 1920a, 1934), the indistinctness of the nucleus of the liver phases, even in stained sections, failure of investigators to cultivate Histomonas from the liver lesions (save only Bishop, 1938), the difficulty in producing infections by oral inoculation with infected liver emulsions—these and other circumstances have justified a certain apprehension in unqualifiedly accepting Histomonas as the sole inciter of entero-hepatitis in turkeys and chicks. Enigk claims that the pathologico-anatomical picture of blackhead as a whole resembles oidiomycosis. The evidence submitted by him deserves further serious consideration. Perhaps, as he suggests, there is a type of blackhead ascribable to Histomonas, and another clinically and pathologically similar one produced by a gemmiparous fungus.

## Trichomonas gallinarum

The views of a number of early workers that enterohepatitis of turkeys was in reality a form of trichomoniasis have already been presented in the discussion of *Histomonas meleagridis*. Most of the main exponents of Histomonas etiology, Smith, Tyzzer, De Volt, and Davis, and others, either mention the presence of Trichomonas at times in the lesions or discuss its possible pathogenicity. De Volt and Davis (1936), particularly, noted that trichomonads were quite frequently associated with histomonads in both liver and cecal lesions and reported that in some cases the former were present in large numbers without the histomonads. They frankly state: "The circumstantial evidence against trichomonads as possible pathogens seems to be increasing rather than diminishing at the present time." Menzani (1933) held *Trichomonas gallinarum* to be but the secondary invader after Blastocystis had damaged the tissues.

Allen (1936, 1941) has openly espoused Trichomonas etiology in enterohepatitis of turkeys without in any way attempting to reflect doubt upon Histomonas etiology. In her earlier paper she considered that the two parasites produced enterohepatitis in poultry "with similar lesions" in the ceca and liver, but in her later work she was able to differentiate among liver lesions due to Trichomonas, Histomonas, and Histomonas and Trichomonas, respectively. Chickens and guinea fowl were also found affected.

The parasite, Allen (1940, 1941) acknowledges, is not Pentatrichomonas as she first reported, but *Trichomonas gallinarum* Martin and Robertson. It grew bacteria-free on Boeck and Drbohlav's (1925) medium consisting of egg slants covered with Locke's solution to which small amounts of liver extract and defibrinated turkey blood were added. The experimental disease produced by oral inoculation resembled the naturally occurring disorder which sometimes terminates fatally.

Liver lesions in trichomonad enterohepatitis appear as granular, cream-colored areas of necrosis with irregular outlines, and are level with or elevated above the liver surface. Size of lesions ranges from pin point to $\frac{3}{4}$ inch. They are notably distinguishable from those in Histomonas livers in that the latter are depressed areas and as a rule larger. The mixed lesions, i.e., containing both Histomonas and Trichomonas, are large, depressed in the centers, and granular and elevated at the border. The ceca of turkeys with trichomoniasis, like those in histomoniasis, contain cheesy cores of blood-stained tissue debris. Trichomonad infection is more inclined to chronicity than the histomonad type. Stabler (1947) considers that confirmatory evidence of the above work is needed. He further suggests that the parasite involved in producing liver lesions may be *T. gallinae* rather than *T. gallinarum*.

Oelson and Allen (1940, 1942) appear effectively to have treated some of the sick birds by fever therapy.

## REFERENCES

Allen, E. A.: 1936. A *Pentatrichomonas* associated with certain cases of enterohepatitis or "black-head" of poultry. Trans. Am. Micr. Soc. 55:315.

———: 1940. A redescription of *Trichomonas gallinarum* Martin and Robertson, 1911, from the chicken and turkey. Proc. Helminth. Soc. Wash. 7:65. '

———: 1941. Macroscopic differentiation of lesions of histomoniasis and trichomoniasis in turkeys. Am. Jour. Vet. Res. 2:214.

Bishop, A.: 1938. *Histomonas meleagridis* in domestic fowls *(Gallus gallus)*. Cultivation and experimental infection. Parasitology 30:181.

Boeck, W. C., and Drbohlav, J.: 1925. The cultivation of *Endamoeba histolytica*. Am. Jour. Hyg. 5:371.

Chester, F. D., and Robin, A.: 1900. Entero-hepatitis or blackhead of fowls. Twelfth Ann. Rep. of Del. Agr. Exper. Sta., p. 60.

Cole, L. J., Hadley, P. B., and Kirkpatrick, W. F.: 1910. Blackhead in turkeys: A study in avian coccidiosis. R. I. Agr. Exper. Sta., Bul. 141:137.

Cram, E. B.: 1927. Bird parasites of the nematode suborders Strongylata, Ascaridata, and Spirurata. U. S. Nat. Mus., Bul. 140.

Curtice, C.: 1907a. The rearing and management of turkeys with special reference to the black-head disease. R. I. Agr. Exper. Sta., Bul. 123:1.

———: 1907b. Further experiments in connection with the blackhead disease in turkeys. R. I. Agr. Exper. Sta., Bul. 124:67.

Cushman, S.: 1893. The production of turkeys. R. I. Agr. Exper. Sta., Bul. 25.

De Volt, H. M., and Davis, C. R.: 1936. Blackhead (infectious enterohepatitis) in turkeys, with notes on other intestinal protozoa. Md. Agr. Exper. Sta., Bul. 392.

Dickinson, E. M.: 1930. Infectious entero-hepatitis in the pea fowl. Jour. Am. Vet. Med. Assn. 76:567.

Drbohlav, J.: 1924. The cultivation of the protozoon of blackhead. Jour. Med. Res. 44:677.

Enigk, K.: 1935. Die Aetiologie der Blinddarm-Leberentzündung der Hühnervögel (Blackhead). Arch. Wiss. u. prakt. Tierheilk. 69:410.

Eriksen, S.: 1925. Blackhead in chicks. Poultry Sci. 4:250.

Glaser, R. W.: 1921. On the cytology and life history of the amoebae. Jour. Parasit. 8:1.

Graybill, H. W.: 1925. Blackhead and other causes of loss of turkeys in California. Univ. Calif. Coll. Agr. Exper. Sta., Circ. 291.

——— and Smith, T.: 1920. Production of fatal blackhead in turkeys by feeding embryonated eggs of *Heterakis papillosa*. Jour. Exper. Med. 31:647.

Hadley, P. B.: 1916a. The role of the flagellated protozoa in infective processes of the intestines and liver. R. I. Agr. Exper. Sta., Bul. 166.

———: 1916b. The avenue and development of tissue-infection in intestinal trichomoniasis. R. I. Agr. Exper. Sta., Bul. 168.

——— and Amison, F. E.: 1911. Further studies in blackhead in turkeys. Zentralbl. f. Bakt. Abt. I., Orig. 58:34.

Higgins, C. H.: 1915. Entero-hepatitis or blackhead in turkeys. Canad. Dept. Agr., Health Animals Branch, Bul. 17.

Jowett, W.: 1911. Blackhead. Infectious entero-hepatitis or typhlo-hepatitis. A disease of young turkeys. Jour. Comp. Path. and Therap. 24:289.

Kaupp, B. F.: 1922. Poultry Diseases. 3rd Ed. Alexander Eger, Chicago.

Milks, H. J.: 1908. A preliminary report on some diseases of chickens. La. Agr. Exper. Sta., Bul. 108:1.

Menzani, C.: 1933. Osservazioni e ricerche su l'entero-epatite infettiva dei tacchini. La Clinica Vet. 56:508.

Mumford, H. W.: 1927. A year's progress in solving some farm problems in Illinois. Ann. Rep., Ill. Agr. Exper. Sta., 1926–27.

Oleson, M. W., and Allen, E. A.: 1940. Treatment of cecal and liver trichomoniasis in turkeys by fever therapy. Proc. Soc. Exper. Biol. and Med. 45:875.

——— and Allen, E. A.: 1942. Fever therapy in the control of cecal and liver trichomoniasis in turkeys. Poultry Sci. 21:120.

Rettger, L. F., and Kirkpatrick, W. F.: 1927. An epidemiological study of blackhead in turkeys. Storrs Agr. Exper. Sta., Bul. 148:285.

Smith, T.: 1895. An infectious disease among turkeys caused by protozoa (infectious entero-hepatitis). Bur. An. Ind., U.S.D.A., Bul. 8:1.

————: 1910. *Amoeba meleagridis.* Science 32:509.

————: 1915. Further investigations into the etiology of the protozoan disease of turkeys known as blackhead, entero-hepatitis, typhlitis, etc. Jour. Med. Res. 33:243.

————: 1917. Some field experiments bearing on the transmission of blackhead in turkeys. Jour. Exper. Med. 25:405.

———— and Graybill, H. W.: 1920a. Epidemiology of blackhead in turkeys under approximately natural conditions. Jour. Exper. Med. 31:633.

———— and Graybill, H. W.: 1920b. Blackhead in chickens and its experimental production by feeding embryonated eggs of *Heterakis papillosa.* Jour. Exper. Med. 32:143.

———— and Smillie, E. W.: 1917. Notes on coccidia in sparrows and their relation to blackhead in turkeys. Jour. Exper. Med. 25:415.

Stabler, R. M.: 1947. *Trichomonas gallinae,* pathogenic trichomonad of birds. Jour. Parasit. 33:207.

Theobald, F. V.: 1907. Parasitic liver disease in poultry. Nat. Poultry Conf., Reading, Off. Rep. 2, p. 181.

Tyzzer, E. E.: 1919. Developmental phases of the protozoon of "blackhead" in turkeys. Jour. Med. Res. 40:1.

————: 1920a. The flagellate character and reclassification of the parasite producing "blackhead" in turkeys—*Histomonas* (gen. nov.) *meleagridis* (Smith). Jour. Parasit. 6:124.

————: 1920b. Observations on the transmission of "blackhead" in turkeys. Jour. Med. Res. 41:219.

————: 1924. The chicken as a carrier of *Histomonas meleagridis* (blackhead): The protozoon in its flagellated stage. Jour. Med. Res. 44:676.

————: 1926. *Heterakis vesicularis* Frölich, 1791: A vector of an infectious disease. Proc. Soc. Exper. Biol. and Med. 23:708.

————: 1927. Entero-hepatitis in turkeys and its transmission through the agency of *Heterakis vesicularis.* Proc. Third World's Poultry Cong.:286.

————: 1934. Studies on Histomoniasis, or "blackhead" infection in the chicken and the turkey. Proc. Am. Acad. Arts and Sciences 69:189.

———— and Collier, J.: 1925. Induced and natural transmission of blackhead in the absence of Heterakis. Jour. Infect. Dis. 37:265.

———— and Fabyan, M.: 1920. Further studies on "blackhead" in turkeys, with special reference to transmission by inoculation. Jour. Infect. Dis. 27:207.

———— and Fabyan, M.: 1922. A further inquiry into the source of the virus in blackhead of turkeys, together with observations on the administration of ipecac and sulphur. Jour. Exper. Med. 35:791.

————, Fabyan, M., and Foot, N. C.: 1921. Further observations on "blackhead" in turkeys. Jour. Infect. Dis. 29:268.

## LEUCOCYTOZOON INFECTIONS[1]

The genus Leucocytozoon is somewhat difficult to define, but it is fundamentally very close to Plasmodium and Haemoproteus. It includes those haemosporidia which appear in the circulating blood as elongate residents of immature red blood corpuscles. There is considerable doubt, however, concerning the true nature of the host cell, and it may indeed be a leukocyte or a lymphocyte. It was formerly stated that the gametocytes were devoid of pigment. Hartman (1929), however, states that the gametocytes of *Leucocytozoon anatis* definitely contain pigment in very small granules scattered throughout the cytoplasm, and O'Roke (1934) gives "pigment granules numerous" as a character of the cytoplasm of nearly mature gametocytes of that species. The host cells of the nearly mature and mature gametocytes become curious elongate membranes, attenuated at the ends, with their deeply staining nuclei drawn out at one margin of the cell along one side of

---

[1]See also Leucocytozoon infections in Chapter 40.

the parasite. The gametocytes themselves are less attenuated and rounded at the ends. The male or microgametocyte, both nucleus and cytoplasm, stains less deeply than the female. The nucleus of the male also tends to have a somewhat larger volume. (See illustration of Leucocytozoon in chapter on Diseases of the Turkey.)

Schizogony occurs in the capillaries of internal organs such as liver, lung, and spleen, perhaps inside endothelial cells. The vectors of Leucocytozoon seem to be principally flies of the genus Simulium. According to O'Roke (1934), the development of *Leucocytozoon anatis* (=*L. simondi*) in *Simulium venustum* may be completed in 5 days or considerably less. In general the cycle in the fly closely parallels that of malaria organisms in mosquitoes.

Birds seem to be the sole hosts of Leucocytozoon. Coatney (1937) has cataloged and host-indexed all species known up to that year. Sixty-eight specific names appear in his list, but there are doubtless some synonyms. Some of the better known representatives are *L. ziemanni* of the little owl (*Athene noctua*), *L. neavei* of the Sudan guinea fowl, *L. anseris* of geese, *L. smithi* of turkeys, *L. simondi* of ducks, and *L. struthionis* of the ostrich. Three species have been described for the common fowl (see Coatney, 1937).

There is cause for little doubt as to the pathogenicity of Leucocytozoon in a number of hosts. O'Roke (1934) noted mortality of 35 per cent in an outbreak among young ducks in Michigan studied by him.

Death usually occurred on the twelfth day. Domestic Peking, Indian runner ducks, wild mallards, and wild black ducks of all ages were susceptible. The parasite concerned was termed *L. anatis* Wickware (1915), but Herman (1938) later showed that the correct name for the duck Leucocytozoon is *L. simondi* Mathis and Leger, 1910, described originally from the teal duck *Querquedula crecca* in Tonkin, China.

Huff (1942) has made certain significant contributions toward clarification of the developmental stages of Leucocytozoon in ducks. The earliest stages consisted of minute ovoid bodies residing in macrophages or extracellularly in liver, spleen, or bone marrow. The more advanced of these already show segregation of their elements in the production of schizonts. In the hepatic cells were noted the "hepatic schizonts," measuring 11 to 18μ. These underwent differentiation into masses called cytomeres; then the latter in turn underwent another and final step in schizogony in becoming transformed into smaller bodies called merozoites. In blood vessels of the heart and spleen, and occasionally also in the liver and intestine, and also extra vascularly, were noted the "megaloschizonts," measuring 60 to 105μ. These differentiate into numerous cytomeres, which in turn differentiate into merozoites with bipolar characteristics. The youngest gametocytes originate from merozoites which invade myelocytes, late polychromatophil erythroblasts, lymphocytes, monocytes, and macrophages. The growth of these

stages, accompanied by distortion of the host, results in the appearances described in the first paragraph.

*L. smithi* Laveran and Lucet, 1905, was first seen in turkeys in eastern United States by Theobald Smith (1895), after whom it is named. It has been reported by Volkmar (1929) as occurring in North Dakota and Minnesota, by Skidmore (1932) in Nebraska, and by Johnson (1942) in Virginia. Other reports indicate its presence in France, Germany, Crimea, and Canada. Skidmore considered *Simulium occidentale* to be the vector concerned in a Nebraska outbreak, while Johnson found S. *nigroparvum* to be the vector in Virginia.

*L. anseris* Knuth and Magdeburg, 1922, was discovered by its describers and by Knuth (1922) in the blood and internal organs of young geese in Germany suffering with a serious and often fatal disease.

*L. bonasae* Clarke (1935) from the ruffed grouse has been shown by Clarke (1935) to be associated with cyclical mortality in the grouse population, and was considered a possible cause of the cycle. Clarke (1938) later worked out an interesting relationship between schizonts and gametocytes with seasons and age of host.

**Therapeutics.** Most of what is known concerning drug treatment has been reviewed by O'Roke (1934) and Coatney and West (1937). Pamakin (plasmochin) proved unsatisfactory, but quinine showed promise if fed for a time before adult gametocytes showed in the blood, but did not affect adult gametocytes. Coatney found that atabrin did attack the adult gametocytes.

**Control.** Control is only to be attained through management. This means, principally, that duck culture should not be attempted in regions where there is running water serving as breeding places for black flies (Simulium). Otherwise, it is necessary to screen the young ducks from the flies, which is difficult. Removing parasitized young and adult ducks from the flock would also prove helpful. Since the young are more susceptible to the disease than the adults, it would help also to control the disease if ducklings were hatched either before or after the main black fly season (see O'Roke, 1934).

## REFERENCES

Clarke, C. H. D.: 1935. Blood parasites of ruffed grouse, *Bonasa umbellus,* and spruce grouse, *Canachites canadensis,* with description of *Leucocytozoon bonasae,* n. sp. Canad. Jour. Res. 12:646.

——: 1938. Organisms of a malarial type in ruffed grouse, with a description of the schizogony of *Leucocytozoon bonasae.* Jour. Wildlife Mgt. 2:146.

Coatney, G. R.: 1937. A catalog and host-index of the genus Leucocytozoon. Jour. Parasit. 23:202.

—— and West, E.: 1937. Some notes on the effect of atabrin on gametocytes of the genus Leucocytozoon. Jour. Parasit. 23:227.

Hartman, E.: 1929. The asexual cycle in *Leucocytozoon anatis.* Jour. Parasit. 15:178.

Herman, C. M.: 1938. *Leucocytozoon anatis* Wickware, a synonym for *L. simondi* Mathis and Leger. Jour. Parasit. 24:472.

Huff, C. G.: 1942. Schizogony and gametocyte development in *Leucocytozoon simondi,* and comparisons with Plasmodium and Haemoproteus. Jour. Infect. Dis. 71:18.

Johnson, E. P.: 1942. Further observations on a blood protozoan of turkeys transmitted by *Simulium nigroparvum* (Twinn). Am. Jour. Vet. Res. 3:214.

Knuth, P.: 1922. Demonstration über in Deutschland gefundene Leucozytozoen der Hausgans. Arch. Schiffs.- u. Trop.-Hyg. 19:185.

—— and Magdeburg, F.: 1922. Ueber ein durch Leucocytozoen verursachtes Sterben junger Gänse. Berliner tierärztl. Wochenschr. 38:359.

O'Roke, E. C.: 1934. A malaria-like disease of ducks caused by *Leucocytozoon anatis* Wickware. Bul. No. 4, Univ. Mich. School Forestry and Conserv.

Skidmore, L. V.: 1932. *Leucocytozoon smithi* infection in turkeys and its transmission by *Simulium occidentale* Townsend. Zentralbl. f. Bakt., Abt. I., Orig. 125:329.

Smith, T.: 1895. An infectious disease among turkeys caused by Protozoa (infectious entero-hepatitis). Bur. An. Ind., U.S.D.A., Bul. 8:1.

Volkmar, F.: 1929. Observations on *Leucocytozoon smithi;* with notes on Leucocytozoa in other poultry. Jour. Parasit. 16:24.

Wickware, A. B.: 1915. Is *Leucocytozoon anatis* the cause of a new disease in ducks? Parasitology 8:17.

## TOXOPLASMA INFECTIONS

To the genus Toxoplasma have been assigned several types of protozoa of uncertain taxonomic affinities that occur widely in the lymphocytes, or indeed in other mononuclear leukocytes, or free in the blood of birds and mammals (Fig. 35.6). They also occur in the parenchymal cells of liver, adrenals, lung, and brain. Wolfson's (1940) review of organisms described as avian Toxoplasma presents some of the difficulties encountered in defining the genus, and should be read by workers interested in Toxoplasma. According to Sabin (1939), however, the capacity to multiply and to produce disease in a variety of hosts, including mammals and birds, are to be regarded as the chief taxonomic characteristics of the group. Morphology as the only guide, he states, may be confusing (see also Wolfson, 1940).

The natural method of transmission remains unknown. Wolfson (1941), however, worked with a strain which she obtained from Dr. James Watt, who in turn first observed it in a guinea pig which had been "injected with some ticks." In the laboratory, Toxoplasma has been transmitted by the injection of peritoneal exudate, blood, and emulsions of various tissues, including brain.

Known hosts for the virulent form, regarded by Sabin as the true Toxoplasma, include the African gondi, rabbits, man, dog, guinea pigs, pigeons, certain wild birds, and canaries. If all these forms are the same species, the correct name is *T. gondii* described by Nicolle and Manceaux in 1908 from the African gondi. The strain from the gondi has been found to infect rabbits, guinea pigs, dogs, mice, moles, pigeons, and Java sparrows (Wenyon, 1926); and Sabin and Olitsky (1937) and Sabin (1939) isolated Toxoplasma from the brain of a guinea pig and showed it to be pathogenic to guinea pigs, rabbits, mice, monkeys, and chickens. Sabin's (1939) account of the transfer of a human strain to mice, thence to rabbits and day-old chicks, further proved the susceptibility of these animals to the human strain. Further, Sabin (1939) showed that strains of human and animal (presumably guinea pig) origin were immunologically identical.

Our present interest in Toxoplasma is that it has been found to occur naturally in pigeons and a number of wild birds, and that it is transmissible to chicks. Two of Sabin's chicks inoculated intracerebrally with infected mouse brain developed nervous symptoms on the fifth and sixth days, respectively. After the natural death of one and the sacrifice of the other on the sixth day, Toxoplasma was demonstrated in their brains microscopically and by passage to other chicks. Two three-week-old chicks inoculated in a similar fashion developed transitory weakness and slight incoordination on the fifth and sixth days, and one showed no symptoms. Four weeks after inoculation,
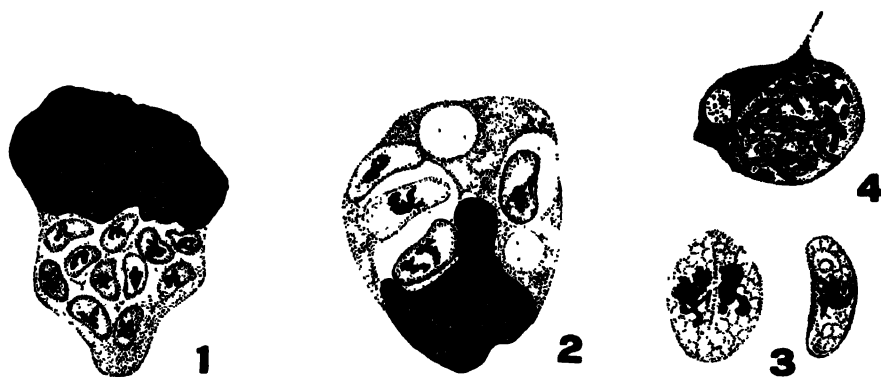


FIG. 35.6. 1, 2—Toxoplasma bodies in lymphoid cells of a ground squirrel. 3—free Toxoplasma in same host, individual on left in division. 4—Toxoplasma in cytoplasm of nerve cell of rabbit. (1–3, after Sassuchin; 4, after Levaditi *et coll.*)

the organisms were demonstrated in the brains of the two chicks by inoculation of mice and other chicks.

Wolfson (1941) transmitted a guinea pig strain to canaries, young ducks, and duck embryos, producing disease and death in most of them. Her work is of further interest in that she found, late in the disease, parasites in the erythrocytes. Guimarães and Meyer (1942) grew Toxoplasma in tissue culture from the spleen, liver, heart, and nerve ganglion of the chick embryo and blood monocytes of the adult chicken. They concluded that binary fission was the only mechanism of reproduction. They verified twisting movements by the parasite, but differing from those of haemogregarines.

There exists in wild birds also a Toxoplasma-like organism which seems not to be transmissible by laboratory injection methods. Such was the form Herman (1937, 1938) found in the blood of the kingbird, catbird, starling, English sparrow, Baltimore oriole, cowbird, red-eyed towhee, savannah sparrow, chipping sparrow, swamp sparrow, and song sparrow on Cape Cod. Herman (1938) was unable to transmit this form from sparrows to canaries, or from English sparrows to chicks, by the conventional laboratory methods.

An authoritative and comprehensive review of mammalian and avian toxoplasmas and the distinction between them was compiled by Manwell,

Coulston, Binckley, and Jones (1945). They include some of the problems associated with the identification, nature, transmission, specificity, host-specificity, and cultivation of this group of microorganisms. Nobrega and Reis (1942) expressed different interpretations regarding the separate nature of mammalian and avian forms.

*Toxoplasma gallinarum* Hepding (1939). Various diseased conditions of the eye were noted by Hepding in many cases of neurolymphomatosis of chickens in Germany. In one case Toxoplasma was observed in the retina. The bodies were elongate, roundish or elliptical, measuring 9.8μ × 7.7μ. Smaller bowed forms, apparently schizonts, measured 3.3μ × 1.6μ, and appeared at the borders of necrotic areas. It was considered that the eye condition was mostly attributable to neurolymphomatosis, with Toxoplasma as a secondary invader.

## REFERENCES

Guimarães, F. N., and Meyer, H.: 1942. Cultivo de "Toxoplasma" Nicolle and Manceaux, 1909, en culturas de tecidos. Rev. Brazil. Biol. 2:123.

Hepding, L.: 1939. Über Toxoplasmen (*Toxoplasma gallinarum* n. sp.) in der Retina eines Huhnes and über deren Beziehung zur Hühnerlähmung. Zeitschr. f. Infektionskr. 55:109.

Herman, C. M.: 1937. Toxoplasma in North American birds and attempted transmission to canaries and chickens. Am. Jour. Hyg. 25:303.

————: 1938. The relative incidence of blood protozoa in some birds from Cape Cod. Trans. Am. Micr. Soc. 57:132.

Manwell, R. D., Coulston, F., Binckley, E. C., and Jones, V.: 1945. Mammalian and avian toxoplasma. Jour. Infect. Dis. 76:1.

Nobrega, P., and Reis, J.: 1942. Idendidade dos toxoplasmos de aves e de mamíferos. Arq. Inst. Biol. São Paulo 13:21.

Sabin, A. B.: 1939. Biological and immunological identity of toxoplasma of animal and human origin. Proc. Soc. Exper. Biol. and Med. 41:75.

———— and Olitsky, P. K.: 1937. Toxoplasma and obligate intracellular parasitism. Science 85:336.

Wenyon, C. M.: 1926. Protozoology. Ballière, Tindall, and Cox, London.

Wolf, A., Cowen, D., and Paige, B.: 1939. Human toxoplasmosis: occurrence in infants as an encephalomyelitis verification by transmission to animals. Science 89:226.

Wolfson, Fruma: 1940. Organism described as avian *Toxoplasma*. Am. Jour. Hyg. 32:88 (Sec. C.).

————: 1941. Mammalian *Toxoplasma* in erythrocytes of canaries, ducks, and duck embryos. Am. Jour. Trop. Med. 21:653.

## HAEMOGREGARINA INFECTIONS

The haemogregarines are members of the suborder Haemogregarinidea, which latter in turn belongs to the order ADELEIDA, of the class **SPOROZOA**. Thus, as Wenyon (1926) points out, the haemogregarines are really of the nature of coccidia rather than gregarines. The forms appearing in the circulating blood, usually in white cells, are usually more or less elongate and enclosed in cysts from which they may emerge when observed in drawn or diluted blood, and move about as peculiar wormlike creatures. This motility is one character which separates them from the nonmotile Toxoplasma. Guimarães and Meyer (1942), however, describe twisting movements in Toxoplasma, but different from those observed in haemogregarines. Their observations were made on Toxoplasma in tissue cultures. The complete

life cycle is unknown for any of the avian haemogregarines, but in the case of certain reptilian and mammalian types the stages inside blood cells are known to be gametocytes without apparent sexual dimorphism. The intermediate host is a blood-sucking invertebrate, such as a leech, tick, or mite. In the invertebrate host fertilization, oocyst formation, and sporozoite formation occur. The vertebrate animal in turn becomes infected by acquiring sporozoites through the bite of the invertebrate or by eating the infected invertebrate, as the case may be.

While admittedly it is necessary to know the whole life cycle of a haemogregarine before it can be placed in a definite family or genus, a number of species have been described. *Haemogregarina adiei* (=*Hepatozoon adiei*) Hoare (1924) from an Indian eagle (species undetermined) appeared in the blood in leukocytes of the large mononuclear type as thick, short rods, rounded at the ends and measuring about $8.5\mu \times 4.5\mu$. The parasites displaced, or were wedged into, the nucleus of the leukocyte. Because of its occurrence within leukocytes and its close morphological resemblance to Hepatozoon in dogs and rats, the parasite was considered by Hoare to be a gametocyte of that genus. Hoare also located schizonts in endothelial cells of the lungs that appeared to be typical of haemogregarines.

Aragão (1911) had previously described five species from Brazilian passerine birds, assigning all of them to the genus Haemogregarina. Later (1933) he defended this procedure. Until more is known about the life cycles it seems advisable to leave all of the avian haemogregarines in the genus Haemogregarina. As a matter of fact, the vector is not known for any of the avian species.

## REFERENCES

Aragão, H. de B.: 1911. Beobachtungen über Hämogregarinen von Vögeln. Mem. Inst. Osw. Cruz 3:54.

————: 1933. Considérations sur les hémogrégarines des oiseaux. Compt. rend. Soc. de biol., Paris 113:214.

Guimarães, F. N., and Meyer, H.: 1942. Cultivo de "*Toxoplasma*" Nicolle and Manceaux, 1909, im culturas de tecidos. Rev. Brazil. Biol. 2:123.

Hoare, C. A.: 1924. *Hepatozoon adiei*, n. sp., a blood parasite of an Indian eagle. Trans. Roy. Soc. Trop. Med. and Hyg. 18:63.

Wenyon, C. M.: 1926. Protozoology. Ballière, Tindall, and Cox, London.

## HAEMOPROTEUS INFECTIONS

The genus Haemoproteus belongs to the family Haemoproteidae which is fundamentally like the family Plasmodiidae, to which the true malaria parasites belong, except that schizogony occurs in endothelial cells of internal organs rather than in circulating blood cells. This fact makes it impossible to transfer the infection by inoculation of infected blood, as can be done with the true malarias. Haemoproteus differs from Leucocytozoon, the other genus in the family Haemoproteidae, principally in the effect of the develop-

ing gametocyte on the parasitized cell. In the case of Leucocytozoon, it becomes spindle-shaped with attenuated ends, while in the case of Haemoproteus it retains approximately its normal shape. The gametocyte of Haemoproteus may push the host-cell nucleus to the side, but it does not flatten it out completely, and the parasite conforms to the shape of the nucleus which it partially encloses "like a halter." The life cycle in general is similar to that of the malaria organisms. The intermediate hosts for the avian infections, in all cases where they are known, are hippoboscid flies. Huff (1942), however, suggests that there may be other dipterous vectors in the case of duck Haemoproteus.

Coatney (1936) published a checklist and host index of the genus Haemoproteus in which appear forty-five specific names, most of which are described from birds. The genus occurs widely in passerine birds, owls, flickers, and woodpeckers, ducks, and other types of birds. Herman (1938a) found 50 per cent of the chipping sparrows on Cape Cod infected. *Haemoproteus lophortyx* O'Roke is a pathogenic parasite of California valley quail (see O'Roke, 1930). Herman (1938b) observed Haemoproteus in a black duck taken at Cape Cod, Massachusetts. A bird may have more than one species; e.g., the mourning dove is the host of both *Haemoproteus maccallumi* and *Haemoproteus sacharovi*. Incidentally, both species have been transferred to the pigeon by means of the vector, *Pseudolynchia maura* (see Huff, 1932). The best known species is *Haemoproteus columbae,* the life cycle of which is well-known (Fig. 35.7). This species is widely distributed throughout the tropical and subtropical world. It occurs in the southern part of the United States, but only sporadically in the northern states because the fly vector, *Pseudolynchia maura,* normally dies out during the winter after chance introduction during the spring and summer. It is ordinarily considered a nonpathogen, but Coatney observed one bird with a very heavy infection that was definitely abnormal. For more detailed information concerning this parasite the reader is referred to Wenyon (1926), Reis and Nobrega (1936), and Coatney (1933).

Atabrin and pamakin (plasmochin), according to Coatney (1935), affect *Haemoproteus columbae* infection. Atabrin inhibits the development of young gametocytes, while pamakin (plasmochin) does not. The latter is parasiticidal to adult gametocytes, however. Neither seems to affect the schizonts.

## REFERENCES

Coatney, G. R.: 1933. Relapse and associated phenomena in the Haemoproteus infection of the pigeon. Am. Jour. Hyg. 18:133.
————: 1935. The effect of atabrin and plasmochin on the Haemoproteus infection of the pigeon. Am. Jour. Hyg. 21:249.
————: 1936. A check-list and host-index of the genus Haemoproteus. Jour. Parasit. 22:88.
Herman, C. M.: 1938a. The relative incidence of blood protozoa in some birds from Cape Cod. Trans. Am. Micr. Soc. 57:132.

————: 1938b. Haemoproteus sp. from the common black duck, *Anas rubripes tristis.* Jour. Parasit. 24:53.

Huff, C. G.: 1932. Studies on Haemoproteus of mourning doves. Am. Jour. Hyg. 16:618.

————: 1942. Schizogony and gametocyte development in *Leucocytozoon simondi* and comparisons with Plasmodium and Haemoproteus. Jour. Infect. Dis. 71:18.

O'Roke, E. C.: 1930. The morphology, transmission, and life-history of *Haemoproteus lophortyx* O'Roke, a blood parasite of the California valley quail. Univ. Calif. Pub. Zool. 36:1.

Reis, J., and Nobrega, P.: 1936. Tratado de Doencas das Aves. São Paulo, Brazil.

Wenyon, C. M.: 1926. Protozoology. Baillière, Tindall, and Cox, London.

## PLASMODIUM INFECTIONS

The true malarial organisms belong to the genus Plasmodium, which in turn is closely related to Haemoproteus and Leucocytozoon. The principal significant difference between Plasmodium and the other two genera is that the asexual stages (schizonts) of the former occur in erythrocytes of the circulating blood, while those of the two latter occur in the internal organs (lung, liver, spleen, kidney, etc.) , presumably inside endothelial cells. As a result, Plasmodium can be transmitted from one susceptible host to another in the laboratory by injection of infected blood from the vessels or heart, while in the case of the other two this procedure will not result in infection because the stages in the blood are solely gametocytes which continue development only in the proper invertebrate host. Like Haemoproteus, Plasmodium contains pigment.



FIG. 35.7. *Haemoproteus columbae.* Pigeon blood. a, b—macrogametocyte in erythrocyte. c—microgametocyte. d, e— normal erythrocyte. (Drake and Jones.)

The life cycle, as is well known, involves two hosts: an intermediate host, a vertebrate, in which asexual multiplication (schizogony) and the formation of immature sexual forms (gametocytes) occur, and a definitive host, presumably always a mosquito, in which maturation of the gametes, fertilization, and sporogony take place. It is of special interest that mammalian malarias are carried by Anopheles mosquitoes, while those of birds are carried by culicine (Culex, Aedes) mosquitoes, although some of the latter have also anopheline vectors. Descriptions of the general life cycle of malaria organisms can be found in almost any textbook in parasitology or protozoology.

Most textbooks, however, do not as yet include the development of Plasmodium from sporozoite to the earliest stages observed in erythrocytes,

the elucidation of which was the contribution of Huff and Coulston (1944). The sporozoite of *P. gallinaceum* enters a "lymphoid macrophage" cell to develop in about 42 hours into a "cryptozoite," or a schizont undergoing schizogony. The merozoites of this generation enter other similar cells to repeat schizogony in another 40 hours (the "metacryptozoites"). Blood infections then ensue, presumably from the merozoites from the fixed tissue stages.

It should be stated at the outset that it is not the writer's intention to discuss comprehensively the malarias of birds, for the literature on the subject is enormous and the number of avian species described more than thirty, although not over half that number may be valid. In fact, Hewitt (1940) has written a book of 228 pages on the subject, and it is not without significance that he states in the preface that on account of cost of publication a certain amount of selection has been made in the choice of subject matter. This book is heartily commended to the reader who is interested in finding extensive bibliography, host records, list of species, key to species, geographical distribution, symptomatology and pathology, therapeutics, laboratory techniques, etc. The catalog and host index of the genus Plasmodium prepared by Coatney and Roudabush (1936) also contains many references to avian species. Since experimental work with bird malarias and publication continues to proceed actively, one could keep abreast of the field only by reading current literature abstracted by *Biological Abstracts.*

Wiselogle (1946) relates the role bird malarias, such as *Plasmodium cathemerium* and *P. relictum* in canaries, *P. gallinaceum* in chicks, and *P. lophurae* in ducks have played in recent investigations for finding better antimalarial drugs.

The following more or less general papers and their bibliographies are also commended to the interested reader: Coatney and Roudabush (1937), Coatney and West (1938), Herman (1938), Huff (1932, 1935a, 1939), Kikuth (1931), Manwell (1935, 1938), Sergent, Sergent, and Catanei (1931), and Wolfson (1941).

Present interest in bird malarias will be confined to species as they affect domestic birds. It is one characteristic of the bird malarias that they are not in general strongly *host specific;* e.g., most (not all) of the species described from wild birds can be grown in the common house canary. The canary has proved invaluable as an experimental host in testing the malaricidal properties of various drugs, but on account of its small size, small blood content, and difficulties encountered in procuring it in numbers, investigators have for some time been looking for suitable domesticated birds as potentially more practical experimental hosts.

### *Plasmodium gallinaceum* Brumpt, 1935

The first important break in the search for a larger avian host came when

Brumpt (1935) discovered *Plasmodium gallinaceum* (Fig. 35.8) in blood smears of an Indo-China chicken, and again the same year in a Ceylon chicken. Because he believed the chicken not to be susceptible to other known avian malarias (see, however, Manwell, 1933), he felt confident he was dealing with a new species. Ducks, guinea fowls, pigeons, turtle doves, quail, buzzards, canaries, sparrows, calfats, and finches were later shown by him to be resistant to infection with *P. gallinaceum,* although chickens of various breeds,· geese, pheasants, partridges, and peacocks were susceptible. The parasite was found by Brumpt to be an extreme pathogen for young chicks, while in adults the infection assumed the chronic form. (See also Coggeshall, 1938.) Since *P. gallinaceum* is a pathogenic microbe of poultry, its importation into the United States is prohibited by quarantine law, save by permission of United States Government authorities. It is, however, being used for experimental work at the Instituto de Salubridad y Enferme-dades Tropicales, Mexico, D. F. So far as is known its natural distribution is still confined to the Orient where Brumpt believes its natural host to be an as yet undetected wild bird with a limited distribution.

Beltran (1941) has recently published a complete summary of the actual state of our knowledge concerning the history, incidence, susceptibility of species, transmission, epidemiology, experimental infection, symptoma-tology and pathology, cytology, life cycle, exoerythrocytic forms, culture, serology and immunology, and chemotherapy of the species. Jacobi (1939) has also studied the pathology of *P. gallinaceum* infection. The discovery of an exoerythrocytic phase, i.e., an asexual developmental cycle in endothelial cells or reticulo-endothelial cells of spleen, brain, liver, and blood, by James and Tate (1938), has attracted a great deal of attention, because it was formerly believed that the increment of malaria parasites in the vertebrate host occurred entirely within erythrocytes. For morphological and other details the reader should consult Brumpt's papers. The known mosquito vectors are *Aedes aegypti, A. albopictus, A. geniculatus,* and possibly *Culex quinquefasciatus* (see Brumpt, 1936a, b; Vargas and Beltran, 1941). Beltran and Larenas (1941) have also demonstrated its transmission by the oral route.

### *Plasmodium lophurae* Coggeshall, 1938

This species was isolated by Coggeshall (1938) from a Borneo fireback pheasant, *Lophura igniti igniti,* at the New York Zoological Park. It is trans-missible to very young chicks, but as a rule produces a moderately severe attack that does not terminate fatally. Only mild infections may be pro-duced in adult fowls, and canaries are not susceptible. The original descrip-tion should be consulted for morphological and other details. Terzian (1941a and b) has recently made an excellent study of the biological characteristics, pathology, and effects of this interesting species in chicks. Laird (1941)

showed that *P. lophurae* can be transmitted from duck to duck through the agency of the mosquito *Aedes albopictus,* and he succeeded in infecting also *Culex restuans* and *Aedes atropalpus. Anopheles quadrimaculatus* can also be infected, at least lightly (cf. Coggeshall, 1941, and Hurlbut and Hewitt,
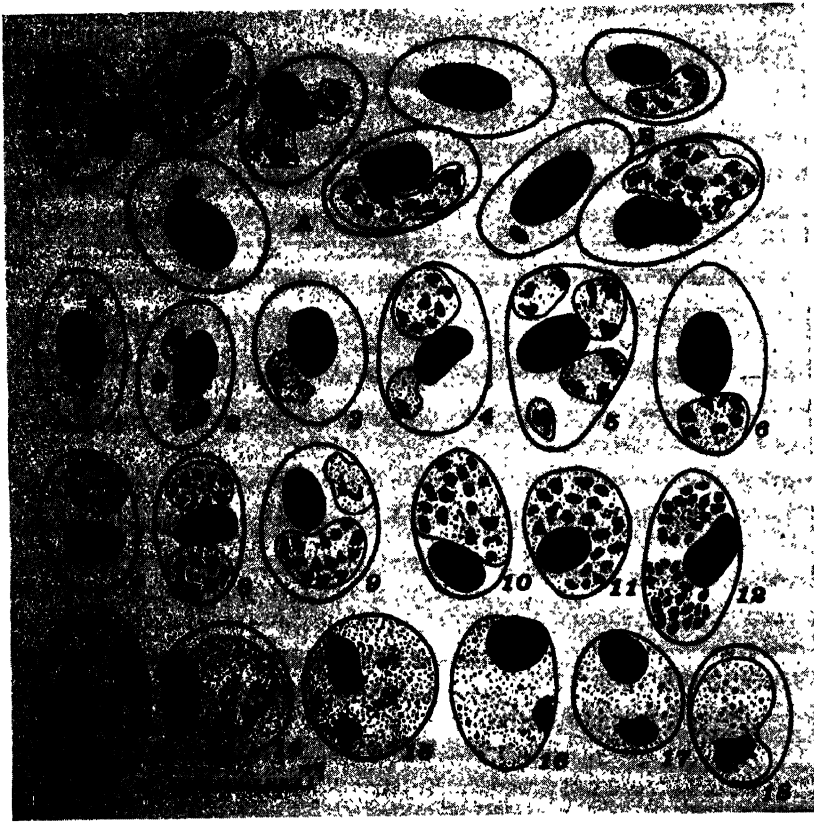


Fig. 35.8. *Plasmodium gallinaceum.* A and B—group of erythrocytes showing alterations in the chromatin of the infected cells. 1–12—simple or multiple infections of the erythrocytes by trophozoites and schizonts in different stages of development. 10, 11, and 12 represent the formation of merozoites; 13, 14, and 15, male gametocytes; 16, 17, and 18, female gametocytes; 18, female gametocyte in an enucleated erythrocyte. (Note: The solid black bodies are the nuclei of the erythrocytes.) (After Brumpt, 1935.)

1941, 1942). Traeger (1942) obtained by selective breeding a strain of *Aedes aegypti* which was more susceptible to *P. lophurae* than the stock from which it had its origin.

Severe infections may be produced in ducks (see Wolfson, 1941). Hewitt's (1942) recent study of host-parasite relationship of *P. lophurae* infection in ducks came to the writer's attention since the foregoing was written. His excellent colored plates of the parasite and types of blood cells

affected by the infection are especially commended to the reader's attention.

### Plasmodium relictum (=P. praecox)

Coatney (1938) obtained strains of *P. relictum* from doves and pigeons that had become naturally infected. The dove strain was extremely pathogenic in pigeons, and canaries and doves were infected with both strains, but the latter infections were light and transitory. Strains were maintained in chicks for as long as 11 days (average, 8.8 days) and in pigeons for 13 days. Subinoculations from chick to chick were carried on for 23 days. Thus, *P. relictum,* a species ordinarily found naturally in many species of wild birds, has produced strains adapted to doves and pigeons, and is capable of growth in chickens as well. Wolfson (1938) also obtained infections in ducks with two other strains of *P. relictum.* Hill (1942) has proved rather conclusively that anemia may be regarded as the cause of death of pigeons in *P. relictum* infections.

### Plasmodium durae Herman, 1941

This species was found in a blood smear of one out of seventy-five domestic turkeys examined in Kenya Colony, British East Africa. It was capable of afflicting young turkeys fatally.

**Other plasmodias.** For further information on avian hosts of avian plasmodia, the reader is referred to Wolfson (1941).

### REFERENCES

Beltran, E.: 1941. Estado actual de nuestros conocimientos acerca del *Plasmodium gallinaceum* Brumpt 1935. Rev. del Inst. Salubr. y Enferm. Trop. 2:95.

—— and Larenas, M. R.: 1941. Produccion di malaria aviar con *Plasmodium gallinaceum* por via oral. Rev. del Inst. Salubr. y Enferm. Trop. 2:87.

Brumpt, E.: 1935. Paludism aviaire: *Plasmodium gallinaceum,* n. sp. de la poule domestique. Compt. Rend. Acad. Sci. 200:783.

——: 1936a. Réceptivité de divers oiseaux domestiques et sauvages au parasite (*Plasmodium gallinaceum*) du paludisme de la poule domestique. Transmission de cet hématozoaire par le moustique *Stegomyia fasciata.* Compt. Rend. Acad. Sci. 203:750.

——: 1936b. Etude expérimentale du *Plasmodium gallinaceum,* parasite de la poule domestique. Transmission de ce germe par *Stegomyia fasciata* et *Stegomyia albopicta.* Ann. Parasit. Hum. et Comp. 14:597.

Coatney, G. R.: 1938. A strain of *Plasmodium relictum* from doves and pigeons infective to canaries and the common fowl. Am. Jour. Hyg. 27:380.

—— and Roudabush, R. L.: 1936. A catalog and host-index of the genus *Plasmodium.* Jour. Parasit. 22:338.

—— and Roudabush, R. L.: 1937. Some blood parasites from Nebraska birds. Am. Midl. Nat. 18:1005.

—— and West, E.: 1938. Some blood parasites from Nebraska birds. II. Am. Midl. Nat. 19:601.

Coggeshall, L. T.: 1938. *Plasmodium lophurae,* a new species of malaria parasite pathogenic for the domestic fowl. Am. Jour. Hyg. 27:615.

——: 1941. Infection of *Anopheles quadrimaculatus* with *Plasmodium cynomolgi,* a monkey malaria parasite, and with *Plasmodium lophurae,* an avian malarial parasite. Am. Jour. Trop. Med. 21:525.

Herman, C. M.: 1938. The relative incidence of blood protozoa in some birds from Cape Cod. Trans. Am. Micr. Soc. 57:132.

——: 1941. *Plasmodium durae,* a new species of malaria parasite from the common turkey. Am. Jour. Hyg. 34:22 (Sec. C).

Hewitt, R.: 1940. Bird malaria. Am. Jour. Hyg., Mon. Series, No. 15, The Johns Hopkins Press, Baltimore.

———: 1942. Studies on the host-parasite relationships of untreated infections with *Plasmodium lophurae* in ducks. Am. Jour. Hyg. 36:6.

Hill, C. McD.: 1942. Anemia as a cause of death in bird malaria. Am. Jour. Hyg. 36:143.

Huff, C. G.: 1932. Further infectivity experiments with mosquitoes and bird malaria. Am. Jour. Hyg. 15:751.

———: 1935a. Natural immunity and susceptibility of culicine mosquitoes to avian malaria. Am. Jour. Trop. Med. 15:427.

———: 1935b. *Plasmodium hexamerium*, n. sp., from the bluebird, inoculable to canaries. Am. Jour. Hyg. 22:274.

———: 1939. A survey of blood parasites of birds caught for banding purposes. Jour. Am. Vet. Med. Assn. 94:615.

——— and Bloom, W.: 1935. A malarial parasite infecting all blood and blood-forming cells of birds. Jour. Infect. Dis. 57:315.

——— and Coulston, F.: 1944. The development of *Plasmodium gallinaceum* from sporozoite to erythrocytic trophozoite. Jour. Infect. Dis. 75:231.

Hurlbut, H. S., and Hewitt, R.: 1941. Sporozoites of *Plasmodium lophurae*, an avian malaria parasite, in *Anopheles quadrimaculatus*. Pub. Health Reps. 56:1336.

——— and Hewitt, R.: 1942. The transmission of *Plasmodium lophurae*, an avian malaria parasite, by *Anopheles quadrimaculatus*. Pub. Health Reps. 57:1891.

Jacobi, L.: 1939. Beiträge zur Pathologie der Infektion des Huhnes mit *Plasmodium gallinaceum* (Brumpt). Arch. f. exper. Path. u. Pharmakol. 191:482.

James, S. P., and Tate, P.: 1938. Exo-erythrocytic schizogony in *Plasmodium gallinaceum* Brumpt, 1935. Parasitology 30:128.

Kikuth, W.: 1931. Immunbiologische und chemotherapeutische Studien an verschiedenen Stämmen von Vogelmalaria. Zentralbl. f. Bakt., etc. Abt. I., Orig. 121:401.

Laird, R. L.: 1941. Observations on mosquito transmission of *Plasmodium lophurae*. Am. Jour. Hyg. 34:163.

Manwell, R. D.: 1933. The behavior of the avian malarias in the common fowl, an abnormal host. Am. Jour. Trop. Med. 13:97.

———: 1935. How many species of avian malaria parasites are there? Am. Jour. Trop. Med. 15:265.

———: 1938. The identification of the avian malarias. Am. Jour. Trop. Med. 18:565.

——— and Goldstein, F.: 1939. Exoerythrocytic stages in the asexual cycle of *Plasmodium circumflexum*. Am. Jour. Trop. Med. 19:279.

——— and Herman, C. M.: 1935. The occurrence of the avian malarias in nature. Am. Jour. Trop. Med. 15:661.

Sergent, Ed., Sergent, Et., and Catanei, A.: 1931. Paludisme des oiseaux. Caractères spécifiques des *Plasmodium aviaires*. Arch. Inst. Past. Alger. 9:399.

Terzian, L. A.: 1941a. Studies on *Plasmodium lophurae*, a malarial parasite in fowls. I. Biological characteristics. Am. Jour. Hyg. 33:1.

———: 1941b. Studies on *Plasmodium lophurae*, a malarial parasite in fowls. II. Pathology and effects of experimental conditions. Am. Jour. Hyg. 33:33.

Trager, W.: 1942. A strain of the mosquito *Aedes aegypti* selected for susceptibility to the avian malarial parasite *Plasmodium lophurae*. Jour. Parasit. 28:457.

Vargas, Luis, and Beltran, E.: 1941. *Culex quinquefasciatus*, a new vector of *Plasmodium gallinaceum*. Science 94:389.

Wiselogle, F. Y.: 1946. A Survey of Antimalarial Drugs. 3 vols. J. W. Edwards, Ann Arbor, Mich.

Wolfson, F.: 1938. The common duck as a convenient experimental host for avian plasmodium. Am. Jour. Hyg. 28:317.

———: 1941. Avian hosts for malaria research. Quart. Rev. Biol. 16:162.

## AEGYPTIANELLA PULLORUM INFECTIONS

This protozoon was first seen by Balfour (1907–14) in Sudanese fowls. His first interpretation of the organism was that it represented intracellular developmental phases of the fowl spirochaete (*Treponema anserinum*) which accompanied the infection. Hindle (1912) and Wenyon (1926) sug-

gested that the intracellular granular bodies represented portions of nuclei extruded into the cytoplasm of the red cell as a result of concomitant spirochaete infection. Balfour (1914) and Carpano (1929) believed that the spirochaete infection and the infection with the granular bodies in the blood
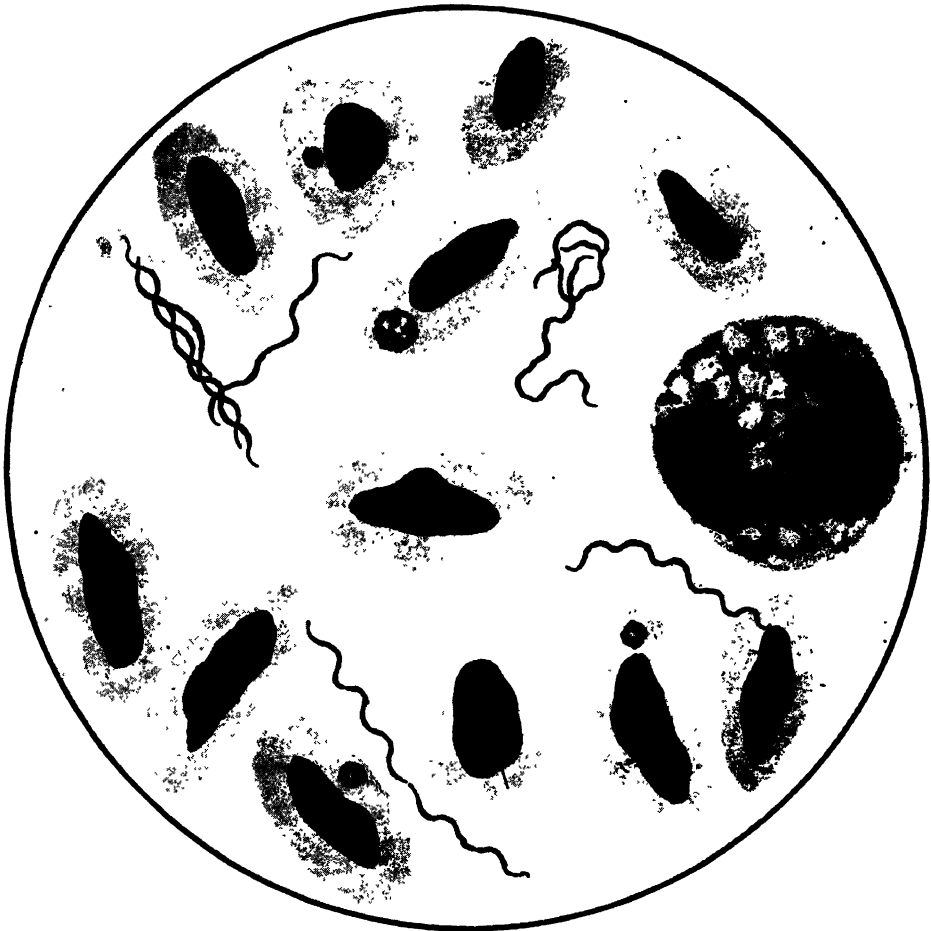


FIG. 35.9. *Aegyptianella pulloıum* and *Spırochaeta gallinaıum.* (Carpano.)

cells were distinct, for either could occur in the absence of the other. Carpano, who has found the organism in both chickens and geese in Egypt, has named the organism *Aegyptianella pullorum,* and considered it a piroplasm (Fig. 35.9) .

Carpano's definition of the genus Aegyptianella is as follows: Protozoa of red cells; of small size; in shape, rounded, oval, or pyriform; not producing pigment; producing no change in size or shape of infected cell; multiplying in circulating blood by schizogony, producing up to 25 minute merozoites.

*A. pullorum* can vary in size from 0.5 to 4.0μ, depending upon the stage of development. Free forms are rarely found in the circulating blood, but may be encountered in the heart, liver, spleen, and bone marrow. In life the parasites may show slow movements of translation. Since the organisms stain with difficulty and the size is small, morphological study is difficult.

**Pathogenicity.** As was previously noted, *A. pullorum* is usually found in association with the fowl spirochaete in Africa. Carpano observed pure infections of both organisms, however, and found that subinoculations with the pure protozoan infection at no time produced spirochaetes.

The protozoan infection may appear in the acute, subacute, or chronic form. Native fowls show the chronic form. Imported birds which become infected are ill for a few days and then succumb. Fowls crossed with foreign strains show the subacute or chronic form.

The symptoms are ruffing of the feathers, anorexia, hyperthermia, immobility, drooping, paralysis of the joints, and often diarrhea. Autopsy shows anemia, splenic tumor, punctiform hemorrhage in the serosa, and sometimes an infiltration of gelatinous hemorrhage in the coronary sulcus.

Brumpt (1930) reported that splenectomy of fowls recovered from heavy infections caused a reappearance of the parasites in the blood. The fowls recovered from this artificially induced relapse.

**Host-specificity.** The infection is found naturally in geese and chickens. Experimental attempts to transmit it to ducks, pigeons, and canaries have been unsuccessful.

**Transmission.** Carpano believes that the frequent presence of Treponema and Aegyptianella in the same bird indicates that *Argas persicus,* the tick transmitter of *T. anserinum,* is also the vector of *A. pullorum.* According to Reis and Nobrega (1936), Coles and Bedford demonstrated carriage by *A. persicus,* although Chaillot and Saunie in 1932 could not confirm it.

**Geographical distribution.** What appears to be this infection has been reported from Egypt, the British and French Sudan, South Africa, and Transcaucasia.

## REFERENCES

Balfour, A.: 1907. A peculiar blood condition, probably parasitic, in Sudanese fowls. Jour. Trop. Med. and Hyg. 10:153.

————: 1911. The role of the infective granule in certain protozoal infections as illustrated by the spirochaetosis of Sudanese fowls. Jour. Trop. Med. and Hyg. 14:113.

————: 1914. Notes on the life-cycle of the Sudan fowl spirochaete. Trans. Seventeenth Internat. Cong. of Med. (London), Sec. 21, Tropical Med. and Hyg., Part 2, pp. 275–78.

Brumpt, E.: 1930. Rechutes parasitaires intenses, dues à la splénectomie, au cours d'infections latentes à Aegyptianella, chez la poule. Compt. Rend. Acad. Sci. 191:1028.

Carpano, M.: 1929. Su di un Piroplasma osservato nei polli in Egitto ("Aegiptianella pullorum") Nota preventiva. Clin. Vet. Milan. 52:339. Also, Bollettino No. 86, Ser. Tec. C Sci., Min. Agric., Cairo, Egypt.

Hindle, E.: 1912. The inheritance of spirochetal infection in *Argas persicus.* Proc. Cambr. Philos. Soc. 16:457.

Reis, J., and Nobrega, P.: 1936. Tratado de Doencas das Aves. São Paulo, Brazil.

Wenyon, C. M.: 1926. Protozoology. Ballière, Tindall, and Cox, London.

## SPIROCHAETE INFECTIONS

*Treponema anserinum* (Sakharoff, 1891) Wenyon, 1926
*Spirochaeta anserina* Sakharoff, 1891
*Spirochaeta gallinarum* Stephens and Christophers, 1904
*Spirochaeta neveuxi* Brumpt, 1909
*Spirochaeta nicolli* Brumpt, 1909
*Spirochaeta granulosa penetrans* Balfour, 1911
*Spirochaeta anatis* Parrot, 1920

This spirochaete was first described from geese in the Caucasus, but it has since been found in the circulating blood of chickens, geese, and ducks in various parts of the world (Fig. 35.9). Turkeys, partridges, crows, sparrows, and other birds, as well as chickens, ducks, and geese, have been experimentally infected by direct blood inoculation. Pigeons have been infected, but in general they are quite resistant. Only light experimental infections have been obtained in rabbits and mice. Tortoises, frogs, and fish also tolerate the spirochaete to a certain degree.

The spirochaete measures from 6 to 30μ in length and from 0.2 to 0.4μ in breadth. The number of undulations varies from 6 to 15. It is actively motile. Reproduction is by transverse division. It was formerly thought that the bodies within erythrocytes, now recognized as Aegyptianella, represented stages in the life cycle of the spirochaete, but the separateness of the two microorganisms is now recognized. Both, however, are transmitted by the fowl tick, or "blue-bug," *Argas persicus*, and both frequently occur within the same fowl. Fowl spirochaetosis has been reported from many parts of the world (Russia, Austria, Hungary, Germany, Bulgaria, Rumania, Greece, Turkey, many parts of Africa, India, East Indies, and Brazil), but so far as the writer knows it has not been reported from the United States, although the "blue-bug" vector occurs in the southern states.

The spirochaete is extremely pathogenic. According to Reis and Nobrega (1936), the period of time between experimental infection by direct inoculation and appearance of the spirochaete in the blood is 2 days; after infection by Argas, 4 to 6 days. Then for 3 days the organisms proceed to multiply actively in the blood, leading up to the crisis with a sharp drop in numbers of spirochaetes. Death may ensue some time after the crisis, sometimes immediately following.

The clinical manifestations of the disease are fever, thirst, ruffed feathers, somnolence, and diarrhea. Later, if they survive, the fowls become weak, emaciated, and anemic. Mortality is about 80 per cent in geese and 60 to 90 per cent in chickens.

Nobrega and Bueno (1945) have discovered that infected fowls may be cured by the intramuscular injection of penicillin. The treated birds not only recovered but were immune to reinfection.

Of an entirely different type may be the spirochaetosis occurring in

turkeys and transmissible to chicks reported from California by Hoffman *et al.* (1946) and Hinshaw and McNeil (1946). In the first place there were no records of Argas or other ticks associated with the affected turkeys. Secondly, the organism is less pathogenic than *Spirochaeta anserina*. Thirdly, the organism does not seem to be associated with *Aegyptianella pullorum*. According to Hinshaw and McNeil (1946) the new spirochaete stains readily with Tunicliff's stain, is soluble in 10 per cent bile, and in 10 per cent saponin, measures 14μ in length, and has an average of six spirals. It can be transmitted to turkeys and chicks by direct inoculation and survives 4 days in adult chickens. The organisms clump in the blood at the peak of infection and become granular prior to death. Mortality is low.

## REFERENCES

Hinshaw, W. R., and McNeil, E.: 1946. Studies on a spirochaete found in the blood of sick turkeys. Jour. Bact. 51:599.

Hoffman, H. A., and Jackson, T. W.: 1946. Spirochetosis in turkeys. Jour. Am. Vet. Med. Assn. 109:481.

———, Jackson, T. W., and Rucker, J. C.: 1946. Spirochetosis in turkeys. Jour. Am. Vet. Med. Assn. 108:329.

Nobrega, P., and Bueno, R. C.: 1945. A ação da penicilina na espiroquetose aviária. Arq. Inst. Biol. São Paulo. 16:15.

Reis, J., and Nobrega, P.: 1936. Tratado de Doenças das Aves. São Paulo, Brazil.

Wenyon, C. M.: 1926. Protozoology. Ballière, Tindall, and Cox, London.

## TRYPANOSOMA INFECTIONS

Trypanosomes are of widespread occurrence in wild birds. Many morphological types have been observed, but there still remain many problems concerning correct species names. In nearly all cases the natural vectors are unknown. Many of the proposed species and their hosts appear in a list by Reis and Nobrega (1936). Only a few of what seem to be definite species are described here.

### *Trypanosoma paddae* Laveran and Mesnil, 1904

This flagellate was originally described from the blood of a Java sparrow (*Padda oryzivora*) sold in a Paris shop. It has a fusiform body, with the anterior end attenuated and accompanying the flagellum to within a short distance of the tip, and the portion posterior to the parabasal body elongated and pointed. Size, 30–40μ by 3–7μ. The nucleus is situated in the middle of the body. The undulating membrane is moderately broad and quite wavy, but so thin it remains uncolored by ordinary blood stains. Multiplication is by binary fission.

Thiroux (1905) infected the following birds by intraperitoneal inoculation: *Serinus meridionalis, S. canarius, Lagonosticta minima, Mariposa phoenicatis,* and *Estrelda cinerea*. Geese, pigeons, English sparrows, and certain other birds were refractory, as were rats, mice, and frogs. More recently MacFie and Thomson (1929) and Manwell and Johnson (1931) have found

trypanosomes occurring naturally in the blood of canary birds in England and the United States, respectively. The English workers believed their flagellate to be *T. paddae,* giving the size as 32–40μ, including flagellum, by 6.0–7.0μ. The Americans considered theirs to be either *T. paddae* or *T. gallinarum,* and gave the size as 30.2μ by 3.62μ. Both sets of workers submitted certain evidence of transmission by the common chicken mite, *Dermanyssus gallinae.*

### *Trypanosoma avium* Danilewsky (1888?)

**Possible synonym.** *T. noctuae* Schaudinn.

According to Laveran, Mesnil, and Nabarro (1907), Danilewsky in a paper published in Russian described the trypanosomes of an owl and roller-bird under the name *Trypanosoma avium.* Laveran (1903) studied a trypanosome of the owl *Syrnium aluco* which he considered the same as that seen by Danilewsky in an owl, but he left the specific status of the form in the roller-bird an open question. Schaudinn's *T. noctuae* of the little owl, *Athene noctua,* is either identical or closely related to *T. avium.* Trypanosomes in wild North American birds have been studied by Novy and Mac-Neal (1905), Coatney and Roudabush (1937), and Coatney and West (1938), and subsequently by certain other workers.

The body is fusiform, with the nucleus at about the middle. Both ends are tapering, and the free flagellum extends some distance from the anterior tip. The posterior end extends a variable distance beyond the parabasal body. The undulating membrane is well developed. Reproduction by binary fission was observed in the blood of the owl by Danilewsky. Schaudinn's declaration that the trypanosome could enter a red cell and become transformed into a Halteridium is no longer tenable.

The transmission of this trypanosome is still unsolved, although Schaudinn (1904) and Woodcock (1914) submitted evidence favoring a mosquito, *Culex pipiens,* as the intermediate host. The development in the mosquito described by Schaudinn is so fanciful that it will not be repeated here. Woodcock found that the blood forms at first underwent multiplication and transformation into crithidias in the digestive tract of the mosquito, and later large and small trypanosome forms appeared. Wenyon (1926), however, is still disposed to regard the method of transmission an open question.

### *Trypanosoma hannai* Pittaluga, 1904

Hanna in 1903 observed a trypanosome in scanty numbers in the blood of domesticated pigeons in India (Fig. 35.10). It was named *Trypanosoma hannai* by Pittaluga (1905). Aragão (1927) also observed the parasite in the blood of a pigeon in Brazil.

Laveran, Mesnil, and Nabarro (1907) give 45 to 60μ for the total length

and 6 to 8μ for the width at the level of the nucleus. The distance from the posterior tip to the parabasal body is 19 to 22μ. The parabasal body is small and situated in a clear vacuole; the undulating membrane is well defined; the free portion of the flagellum is about 7.0μ in length; the nucleus is thin and ribbon-like, and extends across the width of the body; the cytoplasm is either homogeneous or longitudinally striated, and contains granules which are particularly numerous near the posterior end. It may be noted that the accompanying figure by Aragão does not conform to this description in all respects.



FIG. 35.10. *Trypanosoma hannai.* (H. B. Aragão.)

The mode of transmission is still unknown, although Aragão (1927) noted numerous flagellates of the crithidia type in the alimentary tracts of hippoboscid flies, *Lynchia maura,* which had fed upon a pigeon infected with trypanosomes, but none in flies which had fed upon the uninfected birds. Two types of crithidias were noted, the respective dimensions being 40.0μ by 3.0μ and 49.0μ by 1.5μ. Attempts to transmit the infection to clean pigeons by the bite of the fly, by injection of the emulsified intestines of infected flies, or even by blood inoculation were unsuccessful.

## *Trypanosoma gallinarum* Bruce, Hamerton, Bateman, Mackie and Bruce, 1911

Mathis and Leger in 1909 noted the occurrence of relatively short and stumpy trypanosomes (*T. calmetti*), measuring 36.0μ by 4.5μ, in the blood of a common fowl in Tonkin, French Indo-China. Later, Bruce *et coll.* (1911) observed distinctly larger trypanosomes in the centrifuged blood of three fowls in Uganda, which they believed belonged to a different species from the form seen by the French workers. They called the species *Trypanosoma gallinarum.*

In order to find this flagellate it is necessary to centrifuge about 10 cc. of the fowl's blood, and examine under a cover glass several drops of the creamy top layer of leucocytes. In the fresh preparation made as described, the trypanosome remains in one spot, lashes its flagellum about, and performs slow writhing movements of the body and undulating membrane. The length over-all is from 52 to 65μ; breadth, 5 to 7μ. The bandlike nucleus is

situated near the middle of the body. The parabasal body is oblong in shape, from 1 to 1.5μ in length, and situated from 10 to 17μ from the posterior tip of the body. The undulating membrane is broad and thrown into many folds. The free flagellum is from 6 to 10μ long. The body is ribbed longitudinally with parallel lines called myonemes, which extend from the anterior end to the parabasal body.

This trypanosome is inoculable from fowl to fowl by direct blood inoculation, but the numbers in the infected bird remain exceedingly sparse. Monkeys are not susceptible. It grows readily on Novy and MacNeal's blood-agar medium.

The method of transmission is uncertain. Bruce *et coll.*, stated that this trypanosome did not multiply in the intestines of *Glossina palpalis* fed on infected fowls. Duke (1912), however, noted forms of the crithidial type far back in the alimentary tract of tsetse flies (*G. palpalis*) which had fed on infected fowls; but in a note accompanying this report, Muriel Robertson explained that these developmental forms did not indicate that the tsetse fly was the true intermediate host. She stated that she had found small crithidial and trypanosome types in the intestines of an undetermined species of bloodsucking gnats of the family Simuliidae which she had frequently observed feeding upon the blood of Uganda fowls, and favored the view that these would ultimately be found to be the vectors.

## REFERENCES

Aragão, H. de B.: 1927. Eyolution de l'*Haemoproteus columbae* et du *Trypanosoma hannai* dans la *Lynchia maura* Bigot. So. Biol. C. R. 97:827.

Bruce, D., Hamerton, A. E., Bateman, H. R., Mackie, F. P., and Lady Bruce: 1911. *Trypanosoma gallinarum* n. sp. Rep. Sleep. Sickn. Com. Roy. Soc. 11:170.

Coatney, G. R., and Roudabush, R. L.: 1937. Some blood parasites from Nebraska birds. Am. Midl. Nat. 18:1005.

—— and West, E.: 1938. Some blood parasites from Nebraska birds. II. Am. Midl. Nat. 19:601.

Duke, H. L.: 1912. Observations on fowls and ducks in Uganda with relation to *Trypanosoma gallinarum* and *T. gambiense*. Proc. Roy. Soc. (London) 85 B:378–84.

Hanna, W.: 1903. Trypanosoma in birds in India. Quar. Jour. Micr. Sci. 47:433.

Laveran, A.: 1903. Sur un Trypanosome d'une chouette. Compt. rend. Soc. de biol. 55:528.

——, Mesnil, F., and Nabarro, D.: 1907. Trypanosomes and Trypanosomiases. W. T. Keener and Co., Chicago.

MacFie, J. W. S., and Thomson, J. G.: 1929. A trypanosome of the canary, etc. Trans. Roy. Soc. Trop. Med. and Hyg. 23:5–6.

Manwell, R. D., and Johnson, C. M.: 1931. A natural trypanosome of the canary. Am. Jour. Hyg. 14:231.

Novy, F. G., and MacNeal, W. J.: 1905. On the trypanosomes of birds. Jour. Infect. Dis. 2:256.

Pittaluga, G.: 1905. Estudios acerca de los Dipteros y de los parasitos que transmiten al hombre y a los animales domesticos. Rev. Acad. Ciencias, Madrid 3:292, 402, 505.

Reis, J., and Nobrega, P.: 1936. Tratado de Doencas das Aves. São Paulo, Brazil.

Schaudinn, F.: 1903–04. Generations- und Wirtswechsel bei Trypanosoma und Spirochaete. Arb. a. d. kaiserl. Gesundheitsamt. 20:387.

Thiroux, A.: 1905. Recherches morphologiques et expérimentales sur *Trypanosoma paddae* (Laveran et Mesnil). Ann. Inst. Past. 19:65.

Wenyon, C. M.: 1926. Protozoology. Ballière, Tindall, and Cox, London.

Woodcock, H. M.: 1914. Further remarks on the flagellate parasites of Culex. Is there a generic type, Crithidia? Zool. Anz. 44:26.

## INTESTINAL FLAGELLATES
### TRICHOMONAS INFECTIONS

The genus Trichomonas is composed of flagellated protozoa character-ized by the possession of an axial rod, or axostyle, and an undulating mem-brane bordered by a marginal flagellum. The number of flagella at the anterior end may vary according to the species. An attempt has been made to subdivide the genus Trichomonas into the genera Ditrichomonas, Tritri-chomonas, Trichomonas, and Pentatrichomonas, depending on whether the number of free anterior flagella is 2, 3, 4, or 5, but the writer is inclined to ignore this procedure, at least until it can be definitely substantiated that the number of such flagella on individual flagellates does not vary within species. The reader who is desirous of acquainting himself with the detailed structure of a representative of the genus should by all means consult the careful study of *Trichomonas muris* of the rat by Wenrich (1921). Reproduction is principally by binary fission.

There are at least two species of Trichomonas of vital importance in poultry diseases. One, inhabiting the upper digestive tract, is probably cor-rectly known as *T. gallinae,* while the other, inhabiting the lower digestive tract, is *T. gallinarum. T. eberthi* also inhabits the lower digestive tract. Both *T. gallinae* and *T. gallinarum* appear to be definite pathogens though Hinshaw and McNeil (1942) claim to have shown definitely that the diffi-culties usually ascribed to Trichomonas in the lower digestive tract are actually due to Hexamita.

### *Trichomonas gallinarum* Martin and Robertson, 1911

This trichomonad was said by Martin and Robertson (1911) to be one of the commonest flagellates of fowls. It has also been reported from turkeys. According to the original description, the form is more or less spherical, sometimes elongate; size 5.4–7μ in length by 5–6μ in breadth (Fig. 35.11). There were supposed to be four free anterior flagella and a marginal flagel-lum passing down one side of the body attached to a fairly well-developed undulating membrane. All five flagella arose from a blepharoplast complex. Allen (1940), however, has definitely shown that there are five free anterior flagella. The axostyle is slender and sometimes difficult to observe in stained preparations.

This flagellate of the lower digestive tract has been seen by a number of investigators, some of whom ascribed blackhead of turkeys to its activities (see discussion of *Histomonas meleagridis*). The work of Allen, discussed under "Blackhead," indicates it may actually be of great importance in enterohepatitis, though Stabler (1947) suggests the possibility that *T. gal-linae* is the real offender.

Boeck and Tanabe (1926) infected chicks with *T. gallinarum* from turkeys.

*Trichomonas eberthi* Martin and Robertson, 1911

This second trichomonad of the cecum of fowls is said to have a carrot-shaped body measuring about 9μ in length and 4–6μ at the greatest width. Tanabe (1926) gives the usual length as 9–11μ, while Hegner (1929a) gives the measurements as 5.14μ by 2.71μ. There are three free anterior flagella and one marginal flagellum bordering the undulating membrane. (Tanabe says that there are two marginal flagella.) The latter, like that in *T. galli-narum*, ends in a free flagel-lum. The cytostome and axostyle are more developed and conspicuous than in *T. gallinarum*.

Hegner (1929a) infected twelve 2-day-old chicks with *T. eberthi*, and they re-mained infected for at least 102 days. Kotlán (1923) re-ported it for ducks.

*Trichomonas gallinae*
(Rivolta, 1878)

**Synonyms.** *T. columbae* (Rivolta and Delprato, 1880); *T. hepaticum* (Ri-volta, 1878); *T. diversa* Volk-mar, 1930; *T. halli* Yakimoff, 1934.
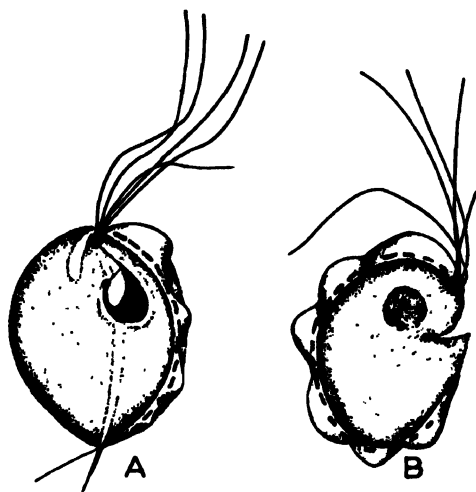


FIG. 35.11. *Trichomonas gallinarum.* A—elon-gated form. B—rounded form. (Allen, Proc. Hel-minth. Soc. Wash.)

According to Stabler (1938a), the turkey and pigeon trichomonad of the anterior digestive tract are identical species, and (Stabler, 1938b) the correct name is *T. gallinae* (Fig. 35.12). The flagellate occurs almost universally in pigeons, and this bird is probably the primary host. The best accounts of the morphology of this species are by Stabler (1941a) and Levine and Brand-ley (1939). A comparison of the figures by these authors with Allen's (1940) figure of *T. gallinarum* brings out several critical points of difference, among them the possession of 4 free anterior flagella in the former and 5 in the latter, the shorter undulating membrane of the former, the definite free flagellum at the end of the marginal flagellum in the latter and its absence in the former, and the more rounded body of the latter in comparison with the more elongate body of the former.

*T. gallinae* in the pigeon occurs in the mouth, esophagus, crop, and in caseous lesions of the liver. The young squabs become infected through the ingestion of their natural food, "pigeon's milk." The pathology in pigeons has been described by Stabler (1947). Mourning doves and other wild birds

may be found infected. In the turkey it appears to be the cause of a severe turkey disease in which it is associated with necrotic ulceration of the crop and lower esophagus, and at times also the proventriculus and upper esophagus (see Volkmar, 1930). Levine and Brandley (1939, 1940) have reported on its pathogenicity in pullets and chicks. Levine, Boley, and Hester (1941) transmitted *T. gallinae* from the chicken to the turkey, quail, canary, and English sparrow with lesions, and to a duckling without lesions. Stabler (1941a) has studied its distribution in wild birds. Treatment is not satisfactory (see Stabler, 1947).
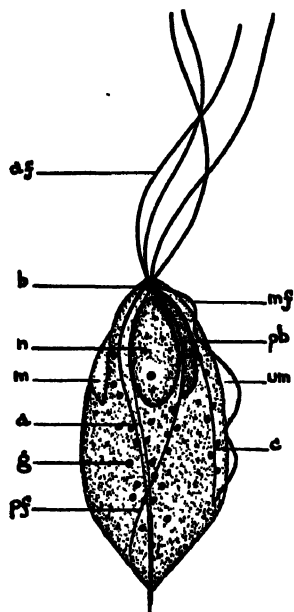


FIG. 35.12. *Trichomonas gallinae,* semidiagrammatic. a—axostyle. af—anterior flagellum. b—blepharoplast. c—costa. g—cytoplasmic granules. m—mouth. mf—marginal filament. n—nucleus. pb—parabasal body. pf—parabasal fibril. um—undulating membrane. (Stabler, Jour. of Morph., of the Wistar Institute of Anatomy and Biology.)

### Trichomonas anatis Kotlán, 1923

**Synonym.** *Tetratrichomonas anatis,* Kotlán, 1923.

Kotlán (1923) made a study of the intestinal flagellates of ducks. His work, except for some random observations, is the only information on this subject available. *T. anatis* has a broadly beet-shaped body which measures 13 to 27μ in length and 8 to 18μ in breadth. There are 4 free anterior flagella of about the same length as the body. The well-developed undulating membrane has a sturdy marginal flagellum along its border.

The habitat is the posterior region of the intestine of *Anas boschas dom.* Kotlán states that massive infections are established only when the mucosa is in a catarrhal condition.

### Trichomonas anseri Hegner, 1929

Hegner (1929a) inoculated 3-day-old chicks with cecal material from a goose containing a very few trichomonads. Some of the chicks became heavily infected. The description of the species is based upon the forms which appeared in the chicks. Hegner finds that *T. anseri* has peculiarities which distinguish it from other trichomonads. The body is oval in shape; size, 6 to 9μ by 3.5 to 6.5μ with a mean of 7.9 by 4.7μ. There are 4 free anterior flagella which arise from 2 blepharoplasts, and a fifth one which forms the border of the undulating membrane and becomes a free lash near the posterior end. The chromatic basal rod is distinct; the axostyle is broad and hyaline and protrudes considerably; the nucleus contains an eccentric karyosome and is otherwise filled with minute chromatin granules. Bacteria are ingested into the cytoplasm through a prominent cytostome.

## *Eutrichomastix gallinarum* Martin and Robertson, 1911

Martin and Robertson (1911) described from the ceca of chickens a form resembling a Trichomonas except that there is no undulating membrane. The body is pyriform, measuring about 5 by 3μ. Four long, free flagella arise from a blepharoplast complex at the anterior end. A single row of deeply-staining granules runs down one side of the body for one-third the body length. The nucleus is oval and contains chromatin scattered more or less irregularly throughout its substance. The habitat of *E. gallinarum* is the ceca. Kotlán (1923) found a similar but larger flagellate in a duck.

### *Chilomastix gallinarum* Martin and Robertson, 1911

This flagellate and its cysts were first reported from fowls by Martin and Robertson (1911), and later by da Fonseca in 1920. The best account is that of Boeck and Tanabe (1926), who also described the process of binary fission in detail (Fig. 35.13).

The body is pyriform, often presenting the slight torsion described for *C. mesnili*. The size is from 11 to 13μ in length and 5 to 6μ in breadth, although smaller and larger forms occur. The cytostome has the shape of a large pouch and extends from the anterior end backwards to almost the middle of the body. The margin of

Fig. 35.13. *Chilomastix gallinarum*, semidiagrammatic illustrating details of morphology. ×5,000. (Boeck and Tanabe, Am. Jour. Hyg.)

the cytostome is supported by right and left supporting fibers as in *C. mesnili*. There are 3 free anterior flagella, and a fourth flagellum vibrates in the cytostome along the border of a small undulating membrane. The action of the undulating membrane is useful in the ingestion of bacteria into the posterior end of the cytostome. Boeck and Tanabe also mention the much-controverted peristomal fibril which Kofoid and Swezy described for *C. mesnili*, but believe that it extends along the base of the embankment of the right side of the groove of the cytostome and ends at the point where the cytostomal flagellum becomes free. The blepharoplasts which give rise to the flagella, right and left supporting fibrils, and peristomal flagella are difficult to count, but there seem to be 4 of them with the relationships to the other organelles shown in the figure. The nucleus is rounded and of the vesicular type.

The unstained cysts resemble those of *C. mesnili* in shape except that they are more slender. They measure 7 to 8.5μ in length and 4.5 to 5.5μ in width. The internal structures are similar to those in the motile form except that

the nucleus has moved to a more central position.

**Location in the host.** The cecum is the normal habitat. The incidence of infection is relatively low.

**Host specificity.** The same species occurs in chickens and turkeys. This statement seems justified on the basis of the morphological identity of the forms in the two birds and the cross-infection experiments of Boeck and Tanabe who infected chicks with the flagellates from the turkey in culture.

### Pleuromonas jaculans

Uribe (1921) observed this flagellate growing in cultures of the fecal material of chickens. The organism answered fairly well to the description of *Pleuromonas jaculans* Perty except that it ingested its food in a ventral indentation rather than on its dorsal side. Uribe states that he found it "in large numbers in the cecal contents of young chickens which had been fed Heterakis material, so that it is evidently adapted to entozoic life."

### Protrichomonas anatis Kotlán, 1923

This species, observed by Kotlán in the duck, has a more or less pear-shaped body measuring 10 to 13μ by 4 to 6μ. There are 3 active flagella which arise from an anterior blepharoplast. It appears as if the axostyle is formed of 2 fibrils which arise from the blepharoplast, pass posteriad, and meet at a point a considerable distance from the caudal tip of the body. The nucleus appears to lie between the fibrils at about the middle of the body.

### Cochlosoma anatis Kotlán, 1923

This curious flagellate has a sharply outlined depression at the anterior end of one side. This concavity, similar to the sucking disc of Giardia, marks the ventral surface. The dorsal surface shows a convexity at the anterior end. The body measures 10 to 12μ by 6 to 7μ. From a blepharoplast on the anterior border of the body there arises a tuft of about 6 flagella, all of which are directed posteriad adjacent to the surface of the body. Two axial fibrils (axostyle ?) arise from a granule near the blepharoplast and traverse the body to beyond the caudal tip. A vesicular nucleus lies in about the center of the body. Ovoid cysts with 4 or more nuclei are formed. Reproduction is either by binary fission of the trophozoite or by multiplication within the cyst (Fig. 35.14).

This organism was found in the feces and intestinal mucus of a duckling which was suffering with coccidiosis of the intestine, and in the ceca of a growing duck. Travis located them also in the cloaca and large intestine of ducks. Kotlán found the same flagellate in *Nyroca ferruginea* and *Fulica atra*. Pathogenicity of the flagellate has not been proved. Travis (1938) observed it in the wild mallard, shoveller, pintail, lesser scaup, and domesticated mallard. Other species were observed in the magpie and Eastern robin.

Kimura in 1934 found a Cochlosoma in Muscovy and white Pekin ducks in California which he named *C. rostratum*. It measured 6–10µ × 3.9–6.7µ. McNeil and Hinshaw (1942) have found it throughout the intestinal tract of turkey poults, and in the region of the cecal tonsil in adults. It occurred in turkeys associated with Hexamita or in combinations of Hexamita and Salmonella. He notes that the true significance of this parasite in turkey poults or ducklings has not been determined.

A severe outbreak of cochlosomiasis, attributed to *C. anatis*, in poults of two to ten weeks on a turkey farm in Scotland was recorded by Campbell
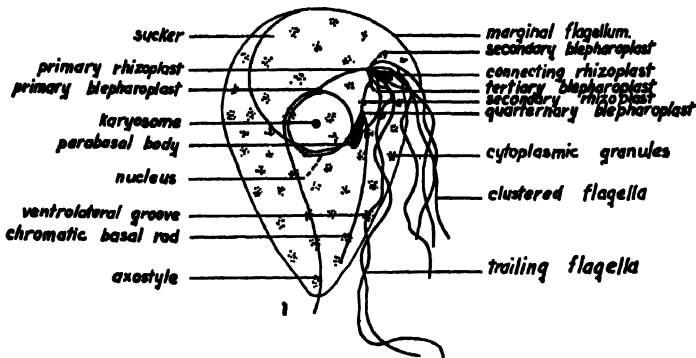


FIG. 35.14. Cochlosoma. Diagrammatic drawing naming structures. (Travis, Jour. of Parasit.)

(1945). The birds were affected with a condition clinically and pathologically indistinguishable from infectious catarrhal enteritis due to Hexamita. The symptoms were as follows: intense thirst, frothy diarrhea, depression, ruffled plumage, drooping head, closed eyes, loss of appetite, weakness, coma, and death. Only 2 or 3 days intervened between the first appearance of symptoms and death. Among the findings at post-mortem was the atonic intestine with dilations or bullae filled with yellow fluid swarming with the jerkily swimming flagellates. The flagellate was revealed in every fresh case. Trichomonas and Hexamita were also usually present, but Cochlosoma predominated.

Carini (1932) described *Cochlosoma pipistrelli* from the anterior intestine of bats of the genus Sternoderma taken at Mogi das Cruzes, São Paulo. Its present interest is that it represented the first occurrence of the genus in other than bird hosts.

## HEXAMITA INFECTION

The genus Hexamita is characterized by the following: (1) Eight free flagella, 6 of which emerge at the anterior end, 3 on each side, and 2 caudal flagella emerging from the body at or near the ends of 2 axostyles or intra-

cytoplasmic portions of the flagella each passing backwards from a granule complex at the anterior end; (2) 2 nuclei, located in the anterior region; (3) absence of the sucker which figures so prominently in the morphology of Giardia. Thus, flagellates of this genus are in reality bilaterally symmetrical. The genus has recently come into prominence in connection with fowl diseases by reason of the work of Hinshaw and his collaborators on infectious catarrhal enteritis of turkeys which makes a strong case for Hexamita etiology (see chapter on Diseases of the Turkey).

### Hexamita meleagridis McNeil, Hinshaw, and Kofoid, 1941

This is the species which, according to Hinshaw, McNeil, and Kofoid (1938a), and Hinshaw and McNeil (1939) is involved in turkey enteritis. In fact, Hinshaw and McNeil go so far as to state that hexamitiasis of turkeys is a disease formerly known as intestinal trichomoniasis, or infectious catarrhal enteritis, and that trichomonads were thought to be the cause of the disorder until it was found in 1938 to be due to Hexamita. The size varies from 6 to 12μ (av. 9μ) in length and 2 to 5μ (av. 3μ) in width. The nuclear membrane is distinct and the karyosomes round and prominent. For further morphological details, see McNeil, Hinshaw, and Kofoid (1941).

In acute enteritis of turkeys *H. meleagridis* is found in large numbers throughout the small intestine and in fewer numbers in the ceca. It was found also in the bursa of Fabricius and, in some poults killed when comatose, in the abdominal cavity and liver (Hinshaw, McNeil, and Kofoid, 1938b). Hinshaw and McNeil (1941) found an incidence of 16.5 per cent in adult turkeys by rectal examination, and 32.4 per cent at autopsy. Thus the incidence is high enough for turkeys to be the most important source of contamination. Chickens, pigeons, ducks, guinea hens, and peafowls were negative for *H. mealagridis*, but *Hexamita columbae* was found in 25 per cent of thirty-two pigeons examined. A species of Hexamita transmissible to poults was found in quail and chickens. English sparrows, linnets, butcher birds, towhees, hawks, an owl, a kingbird, and a crow were found negative. The infection persisted for at least twenty-two weeks in chicks inoculated from turkeys, though symptoms of infectious catarrhal enteritis were not produced (McNeil and Hinshaw, 1941a).

### Hexamita columbae Nöller and Buttgereit, 1923

**Synonym.** *Octomitus columbae* Nöller and Buttgereit, 1923.

The meagre description of this flagellate was supplied in a brief note by Nöller and Buttgereit (1923). The body measures 5 to 9μ by 2.5 to 7μ, and is said to be plumper than that of *H. muris* and very seldom pointed behind. The nuclei are rounded. The organism occurred in great masses from the stomach to the anus of a pigeon stricken with intestinal catarrh.

McNeil and Hinshaw (1941b) have reported four cases of Hexamita in stricken pigeons in California. They report it was not possible for them to

infect turkey poults with *H. columbae,* nor did they find *H. meleagridis* in the pigeons on ranches where the turkeys were heavily parasitized with that species.

## · *Hexamita* sp.

Kotlán (1923) recorded the presence of *"Hexamitus intestinalis* (?) " in the intestinal mucus of *Anas boschas dom.* The description of this form was meagre, and no figures were given. The shape of the body was that of an elongated pear, or ovoid. The size was 10 to 13µ in length by 4 to 5µ in breadth. The 8 flagella, arising from several basal granules, were of considerable length; the 2 axostyles either traversed the length of the body parallel to each other or crossed; 2 oval nuclei lay at the anterior end of the body; no cytostome was observed.

*Note.* Collier and Boeck (1926) produced temporary infections of the ceca of chicks with *Embadomonas cuniculi* of the rabbit. Hegner (1929a, 1929b) achieved more or less success in infecting the ceca of chicks with the following flagellates from foreign hosts: *Trichomonas hominis, T. buccalis, T. anthropopitheci, Trichomonas* sp. from rumen of sheep, *T. suis, T. caviae, T. muris, T. parva, T. cynomysi, T. anatis, T. anseri, T. oti, T. floridane, T. ortyxis, Chilomastix mesnili, C. bittencourti, C. intestinalis, Giardia lamblia* (excystment only), *G. muris* (excystment only), *Embadomonas caviae.* "*T. parva*" from the rat was perhaps the seemingly ubiquitous "Pentatrichomonas" found likewise in man.

### REFERENCES

Allen, E. A.: 1936. A pentatrichomonas associated with certain cases of enterohepatitis or "black-head" of poultry. Trans. Am. Micr. Soc. 55:315.

————: 1940. A redescription of *Trichomonas gallinarum* Martin and Robertson, 1911, from the chicken and turkey. Proc. Helminth. Soc. Wash. 7:65.

Boeck, W. C., and Tanabe, M.: 1926. *Chilomastix gallinarum.* morphology, division, and cultivation. Am. Jour. Hyg. 6:319.

Campbell, J. G.: 1945. An infectious enteritis of young turkeys associated with *Cochlosoma* sp. Vet. Jour. 101:255.

Carini, A.: 1932. Sobre um flagelado do genero Cochlosoma encontrado no intestino de morcegos. Arq. d. Biol., Ano XXIII, Num. 217 (separate with page number).

Collier, J., and Boeck, W. C.: 1926. The morphology and cultivation of *Embadomonas cuniculi* n. sp. Jour. Parasit. 12:131.

Gierke, A. G., and Hinshaw, W. R.: 1936. Mortality in young turkeys associated with tricho-moniasis. Jour. Am. Vet. Med. Assn. 88:76.

Hawn, M. C.: 1937. Trichomoniasis of turkeys. Jour. Infect. Dis. 61:184.

Hegner, R. W.: 1929a. Transmission of intestinal protozoa from man and other animals to parasite-free fowls. Am. Jour. Hyg. 9:529.

————: 1929b. The infection of parasite-free chicks with intestinal protozoa from birds and other animals. Am. Jour. Hyg. 10:33.

Hinshaw, W. R., McNeil, E., and Kofoid, C. A.: 1938a. The relationship of *Hexamita* sp. to an enteritis of turkey poults. Cornell Vet. 28:281.

————, McNeil, E., and Kofoid, C. A.: 1938b. The presence and distribution of *Hexamita* sp. in turkeys in California. Jour. Am. Vet. Med. Assn. 93:160.

———— and McNeil, E.: 1939. Infectious catarrhal enteritis of turkeys. Turkey Talk 1:5, 7. 21

———— and McNeil, E.: 1941. Carriers of *Hexamita meleagridis.* Am. Jour. Vet. Res. 2:453.

———— and McNeil, E.: 1942. Hexamitiases in turkeys. Turkey World 17: (May) 16.

cytoplasmic portions of the flagella each passing backwards from a granule complex at the anterior end; (2) 2 nuclei, located in the anterior region; (3) absence of the sucker which figures so prominently in the morphology of Giardia. Thus, flagellates of this genus are in reality bilaterally symmetrical. The genus has recently come into prominence in connection with fowl diseases by reason of the work of Hinshaw and his collaborators on infectious catarrhal enteritis of turkeys which makes a strong case for Hexamita etiology (see chapter on Diseases of the Turkey).

### *Hexamita meleagridis* McNeil, Hinshaw, and Kofoid, 1941

This is the species which, according to Hinshaw, McNeil, and Kofoid (1938a), and Hinshaw and McNeil (1939) is involved in turkey enteritis. In fact, Hinshaw and McNeil go so far as to state that hexamitiasis of turkeys is a disease formerly known as intestinal trichomoniasis, or infectious catarrhal enteritis, and that trichomonads were thought to be the cause of the disorder until it was found in 1938 to be due to Hexamita. The size varies from 6 to 12µ (av. 9µ) in length and 2 to 5µ (av. 3µ) in width. The nuclear membrane is distinct and the karyosomes round and prominent. For further morphological details, see McNeil, Hinshaw, and Kofoid (1941).

In acute enteritis of turkeys *H. meleagridis* is found in large numbers throughout the small intestine and in fewer numbers in the ceca. It was found also in the bursa of Fabricius and, in some poults killed when comatose, in the abdominal cavity and liver (Hinshaw, McNeil, and Kofoid, 1938b). Hinshaw and McNeil (1941) found an incidence of 16.5 per cent in adult turkeys by rectal examination, and 32.4 per cent at autopsy. Thus the incidence is high enough for turkeys to be the most important source of contamination. Chickens, pigeons, ducks, guinea hens, and peafowls were negative for *H. mealagridis*, but *Hexamita columbae* was found in 25 per cent of thirty-two pigeons examined. A species of Hexamita transmissible to poults was found in quail and chickens. English sparrows, linnets, butcher birds, towhees, hawks, an owl, a kingbird, and a crow were found negative. The infection persisted for at least twenty-two weeks in chicks inoculated from turkeys, though symptoms of infectious catarrhal enteritis were not produced (McNeil and Hinshaw, 1941a).

### *Hexamita columbae* Nöller and Buttgereit, 1923

**Synonym.** *Octomitus columbae* Nöller and Buttgereit, 1923.

The meagre description of this flagellate was supplied in a brief note by Nöller and Buttgereit (1923). The body measures 5 to 9µ by 2.5 to 7µ, and is said to be plumper than that of *H. muris* and very seldom pointed behind. The nuclei are rounded. The organism occurred in great masses from the stomach to the anus of a pigeon stricken with intestinal catarrh.

McNeil and Hinshaw (1941b) have reported four cases of Hexamita in stricken pigeons in California. They report it was not possible for them to

infect turkey poults with *H. columbae,* nor did they find *H. meleagridis* in the pigeons on ranches where the turkeys were heavily parasitized with that species.

## *Hexamita* sp.

Kotlán (1923) recorded the presence of *"Hexamitus intestinalis* (?) " in the intestinal mucus of *Anas boschas dom.* The description of this form was meagre, and no figures were given. The shape of the body was that of an elongated pear, or ovoid. The size was 10 to 13μ in length by 4 to 5μ in breadth. The 8 flagella, arising from several basal granules, were of considerable length; the 2 axostyles either traversed the length of the body parallel to each other or crossed; 2 oval nuclei lay at the anterior end of the body; no cytostome was observed.

*Note.* Collier and Boeck (1926) produced temporary infections of the ceca of chicks with *Embadomonas cuniculi* of the rabbit. Hegner (1929a, 1929b) achieved more or less success in infecting the ceca of chicks with the following flagellates from foreign hosts: *Trichomonas hominis, T. buccalis, T. anthropopitheci, Trichomonas* sp. from rumen of sheep, *T. suis, T. caviae, T. muris, T. parva, T. cynomysi, T. anatis, T. anseri, T. oti, T. floridane, T. ortyxis, Chilomastix mesnili, C. bittencourti, C. intestinalis, Giardia lamblia* (excystment only) , *G. muris* (excystment only) , *Embadomonas caviae. "T. parva"* from the rat was perhaps the seemingly ubiquitous "Pentatrichomonas" found likewise in man.

### REFERENCES

Allen, E. A.: 1936. A pentatrichomonas associated with certain cases of enterohepatitis or "blackhead" of poultry. Trans. Am. Micr. Soc. 55:315.

——: 1940. A redescription of *Trichomonas gallinarum* Martin and Robertson, 1911, from the chicken and turkey. Proc. Helminth. Soc. Wash. 7:65.

Boeck, W. C., and Tanabe, M.: 1926. *Chilomastix gallinarum,* morphology, division, and cultivation. Am. Jour. Hyg. 6:319.

Campbell, J. G.: 1915. An infectious enteritis of young turkeys associated with *Cochlosoma* sp. Vet. Jour. 101:255.

Carini, A.: 1932. Sobre um flagelado do genero Cochlosoma encontrado no intestino de morcegos. Arq. d. Biol., Ano XXIII, Num. 217 (separate with page number) .

Collier, J., and Boeck, W. C.: 1926. The morphology and cultivation of *Embadomonas cuniculi* n. sp. Jour. Parasit. 12:131.

Gierke, A. G., and Hinshaw, W. R.: 1936. Mortality in young turkeys associated with trichomoniasis. Jour. Am. Vet. Med. Assn. 88:76.

Hawn, M. C.: 1937. Trichomoniasis of turkeys. Jour. Infect. Dis. 61:184

Hegner, R. W.: 1929a. Transmission of intestinal protozoa from man and other animals to parasite-free fowls. Am. Jour. Hyg. 9:529.

——: 1929b. The infection of parasite-free chicks with intestinal protozoa from birds and other animals. Am. Jour. Hyg. 10:33.

Hinshaw, W. R., McNeil, E., and Kofoid, C. A.: 1938a. The relationship of *Hexamita* sp. to an enteritis of turkey poults. Cornell Vet. 28:281.

——, McNeil, E., and Kofoid, C. A.: 1938b. The presence and distribution of Hexamita sp. in turkeys in California. Jour. Am. Vet. Med. Assn. 93:160.

—— and McNeil, E.: 1939. Infectious catarrhal enteritis of turkeys. Turkey Talk 1:5, 7, 21.

—— and McNeil, E.: 1941. Carriers of *Hexamita meleagridis.* Am. Jour. Vet. Res. 2:453.

—— and McNeil, E.: 1942. Hexamitiases in turkeys. Turkey World 17: (May) 16.

Kotlán, A.: 1923. Zur Kenntnis der Darmflagellaten aus der Hausente und anderen Wasservögeln. Zentralbl. Bakt., Abt. I. Orig. 90:24.

Levine, N. D., Boley, L. E., and Hester, H. R.: 1941. Experimental transmission of *Trichomonas gallinae* from the chicken to other birds. Am. Jour. Hyg. 33:23.

―――― and Brandley, C. A.: 1939. A pathogenic Trichomonas from the upper digestive tract of chickens. Jour. Am. Vet. Med. Assn. 95:77.

―――― and Brandley, C. A.: 1940. Further studies on the pathogenicity of *Trichomonas gallinae* for baby chicks. Poultry Sci. 19:205.

Martin, C. H., and Robertson, M.: 1911. Further observations on the cecal parasites of fowls, with some reference to the rectal fauna of other vertebrates. Part I. Quart. Jour. Micr. Sci. 57:53.

McNeil, E., and Hinshaw, W. R.: 1941a. Experimental infection of chicks with *Hexamita meleagridis*. Cornell Vet. 31:345.

―――― and Hinshaw, W. R.: 1941b. The occurrence of Hexamita (*Octomitus columbae*) in pigeons in California. Jour. Parasit. 27:185.

―――― and Hinshaw, W. R.: 1942. *Cochlosoma rostratum* from the turkey. Jour. Parasit. 28:349.

――――, Hinshaw, W. R., and Kofoid, C. A.: 1941. *Hexamita meleagridis* sp. nov. from the turkey. Am. Jour. Hyg. 34:71.

――――, Platt, E. D., and Hinshaw, W. R.: 1939. Hexamita sp. from quail and from chicken partridges. Cornell Vet. 29:330.

Nöller, W., and Buttgereit, F.: 1923. Über ein neues parasitisches Protozoon der Haustaube. Zentralbl. f. Bakt., Abt. I, Ref. 75:239.

Stabler, R. M.: 1938a. The similarity between the flagellate of turkey trichomoniasis and *T. columbae* in the pigeon. Jour. Am. Vet. Med. Assn. 93:33.

――――: 1938b. *Trichomonas gallinae* (Rivolta, 1878), the correct name for the flagellate in the mouth, crop, and liver of the pigeon. Jour. Parasit. 24:553.

――――: 1941a. The morphology of *Trichomonas gallinae* (=*columbae*). Jour. Morph. 69:501.

――――: 1941b. Further studies on trichomoniasis in birds. The Auk 58:558.

――――: 1947. *Trichomonas gallinae*, pathogenic trichomonad of birds. Jour. Parasit. 33:207.

Tanabe, M.: 1926. Morphological studies on *Trichomonas*. Jour. Parasit. 12:120.

Travis, B. V.: 1938. A synopsis of the flagellate genus Cochlosoma Kotlán, with the description of two new species. Jour. Parasit. 24:343.

Uribe, C.: 1921. A common infusion flagellate occurring in the caecal contents of the chicken. Jour. Parasit. 8:58.

Volkmar, F.: 1930. *Trichomonas diversa* n. sp. and its association with a disease of turkeys. Jour. Parasit. 17:85.

Wenrich, D. H.: 1921. The structure and division of *Trichomonas muris* (Hartmann). Jour. Morph. 36:119.

## PARASITIC AMOEBAE
### *Entamoeba gallinarum* Tyzzer, 1920

This amoeba was described by Tyzzer (1920) from the cecal excrement of both young turkeys and the common fowl. The trophozoites measure from 9 to 25μ in diameter; average size, 16 to 18μ. They are continuously and actively motile at room temperature. Pseudopod formation is said by Tyzzer to be gradual rather than eruptive in character, but Hegner (1929b) finds that it is almost as explosive as in *E. histolytica*. The ectoplasm is differentiated from the endoplasm. The latter stains intensely and usually contains a variety of inclusions such as cell fragments, flagellates, amoebae of the genus Endolimax, and other material from the cecal contents. Strangely enough, Tyzzer states that bacteria are not utilized as food by this species. The nucleus is spherical, and measures from 3 to 5μ across. A dense layer of chromatin is closely applied to the nuclear membrane. Tyzzer states that the endosome is centrally located, but in his figures he shows it in an eccentric position.

The cysts are eight-nucleate when mature, but the immature quadri-nucleate forms are often encountered. The nuclei are said to be distributed and around a large, deeply-staining central mass. The cysts are spheroidal and have an average size of 12 by 15μ.

This amoeba is not known to affect the host adversely in life, but within a short time after death the organisms migrate through the tissue and can be found in large numbers throughout the cecal mucosa and submucosa. Under these conditions the parasite may also ingest epithelial cells.

If it should eventually be determined that this Entamoeba is identical with that found in the red grouse, the correct name would be *E. lagopodis* Fantham, 1910. Although only four-nucleate cysts of the latter species were noted by Fantham, it is not unlikely that the mature cysts possess eight nuclei. What seems to be *E. gallinarum* was reported from guinea fowls by Hegner (1929b).

<center>*Endolimax gregariniformis* Tyzzer, 1920</center>

**Synonyms.** *Pygolimax gregariniformis* Tyzzer, 1920; *Endolimax janisae* Hegner, 1926.

This small amoeba was found by Tyzzer (1920) in the ceca of diseased and normal turkeys, is ovoid in shape and with a posterior protuberance measures, according to Tyzzer, from 3.9 to 13.3μ; average size, 8.75 by 5.3μ. The movement is quite sluggish at room temperature. The ectoplasm is not noticeably differentiated from the endoplasm. The cytoplasm shows a variable number of food vacuoles in which bacteria or other bodies may be enclosed. The nucleus shows a large, deeply-staining, centrally situated kary-osome separated from an achromatic membrane by a clear space. The diame-ter of the nucleus is from 1.5 to 2.0μ; that of the karyosome, from 0.8 to 1.3μ. Uninucleate rounded forms resembling cysts were also found. Tyzzer ob-served both intranuclear and cytoplasmic parasites of the motile amoebae. The former were coccus-like bodies, and the latter appeared to be minute colonies of Sphaerita.

A similar amoeba was described from the common fowl by Hegner (1926) under the name *Endolimax janisae*.

Tyzzer was able to transmit this species readily to young chicks by feeding small amounts of the feces of the infected adult birds. Myriads of the small amoebae were present in the cecal discharges of the chicks 4 days after the inoculation. Hegner (1929b) found what seems to be the same amoeba in guinea fowls. He also inoculated chicks per os and per rectum with the cecal contents of a goose. There appeared in the ceca of the chicks large numbers of an *Endolimax amoeba*. They measured 4.5 to 11μ in length by 4.5 to 8μ in breadth; average size, 7.8 by 6.2μ. Cysts were observed, but no measurements were given. Hegner is uncertain whether this form is co-specific with *E. gregariniformis* of chickens or represents a new species. In addition, the

same investigator noted an Endolimax in cecal material of a duck, and succeeded in cultivating it in chicks.

### Endolimax numidae Hegner, 1929

This small species described from guinea fowls in the United States differs from *Endolimax gregariniformis* in its smaller average size which is given as 4.2 by 3.4μ. The mature cyst is tetranucleate.

### Entamoeba anatis Fantham, 1924

Fantham (1924) in Africa found this histolytica-like Entamoeba in the feces of a duck which had died of an acute enteritis. The appearance, structure, and size of the trophozoites and the presence of erythrocytes in the endoplasm all indicated a similarity to *E. histolytica* of man. In addition, the cysts were spherical or subspherical, uninucleate and tetranucleate, thin-walled, possessed of thin, needle-like, chromatoid bars, and measured 13 by 14μ. A further study should be made of this unusual organism.

### Entamoeba sp.

Hegner found in chicks previously inoculated with cecal material from the duck an Entamoeba resembling that from the guinea fowl. The nucleus, however, was spherical in shape and not irregular, and the chromatin was massed in conspicuous clumps on the membrane. The motile forms ranged from 14 to 22.5μ in diameter; average, 17.78μ. No cysts were observed.

#### OTHER AMOEBAE

Hegner (1929a) was unsuccessful in transmitting per rectum or per os the human amoebae *E. coli, Endolimax nana,* and *Iodamoeba williamsi* to chicks from 3 to 18 days of age. *Entamoeba histolytica, E. muris,* and *E. caviae,* however, persisted in the cecum up to 2 days, 5 days, and 20 hours, respectively. Later (1929b) he found that an Endolimax from the stomach of sheep could establish itself in the ceca of chicks for as long as 21 days. The cecal discharges contained both trophozoites and cysts.

Hegner (1929b) also succeeded in infecting chicks with the following entozoic amoebae of birds: *Entamoeba* sp. and *Endolimax numidae* of guinea fowls, *Entamoeba anatis* (?) of the duck, *Endolimax* sp. from the goose, and *Endolimax* sp. supposedly from the feces of the screech owl.

#### REFERENCES

Fantham, H. B.: 1924. Some parasitic protozoa found in South Africa: VII. So. African Jour. Sci. 21:435.

Hegner, R. W.: 1926. *Endolimax caviae* n. sp. from the guinea-pig and *Endolimax janisae* n. sp. from the domestic fowl. Jour. Parasit. 12:146.

———: 1929a. Transmission of intestinal protozoa from man and other animals to parasite-free fowls. Am. Jour. Hyg. 9:529.

———: 1929b. The infection of parasite-free chicks with intestinal protozoa from birds and other animals. Am. Jour. Hyg. 10:33.

Tyzzer, E. E.: 1920. Amoebae of the caeca of the common fowl and of the turkey. Jour. Med. Res. 41:199.

## CHAPTER THIRTY-SIX

## DISEASES OF THE DIGESTIVE SYSTEM

*By* A. J. Durant and H. C. McDougle, *Department of Veterinary Science,
University of Missouri, Columbia, Missouri*

☆　☆　☆

Anomalies or changes which are not described or discussed under specific diseases are included in this chapter.

### THE BEAK

The beak of the fowl is a hard, prehensile organ used primarily for taking up food and is not subject to many anomalies that can be classified as diseases. The most common change is the condition known as "scissors bill" (Fig. 36.1), in which the premaxilla and mandible are not in apposition, but are crossed. This appears to be a congenital condition of the mandible and premaxilla making it difficult for birds to pick up their food normally. Thus, they fail to develop at the same rate as their companions, and the result is usually a stunted individual. Extreme cases should be destroyed.

Another condition also probably congenital is that in which the upper or lower part of the beak may be much shorter than its opposing structure. This tends to make it difficult for the bird to take in its food and will result in a stunted individual. If the condition is sufficiently severe, the bird should be destroyed as soon as it is discovered.

Parrots may develop horny growths on the beak when affected with tuberculosis; in fact, the condition is rather common (Fig. 36.3).

A condition in which feed became packed between the horny beak and underlying tissues and which resulted in "pressure necrosis" and destruction of the underlying tissues was of common occurrence in the past. Improved feeding practices probably account for its rare occurrence at present. At the time when this condition was quite prevalent among flocks, it was thought that the addition of bran to the diet, by making the ration coarser, eliminated the trouble.

Because of the nature of the beak, fractures of its parts are rare.

In sinus infection (sinusitis) with solid pus formation in the sinuses, the position of the mandible may be displaced and the bird unable to close its beak.

In young birds cockleburs have been known to lodge between the hori-

zontal parts of the mandible. They have likewise been found lodged in the esophagus, glandular stomach, and gizzard. Clefts or openings in the oral cavity are rare; one case was observed in a pelican which had had the pouch slit open, probably due to an accident. When fish were taken into the mouth they would drop out as fast as taken in. The bird was in an advanced stage of starvation. The cleft was first noticed when the tongue appeared through the opening. The question is raised as to whether it was of traumatic or congenital origin.

A bird showing an unusual deformity (Fig. 36.2) was noticed among a large group of experimental birds. The tongue, nearly normal in size, protruded through a complete fistula in the posterior floor of the oral cavity. The anterior two-thirds of the tongue was dehydrated, yet the transverse row of papillae and the hyoid were normal.

A normal horny or transparent formation



FIG. 36.1. "Scissors" bill in a young poult. This turkey was hatched with this condition.

sometimes appears on the end of the tongue, and its removal has been suggested for relief. It is referred to as a "pip" by laymen. No other changes are likely to occur in the mouth and beak of birds other than those produced by specific disease, such as the internal (diphtheritic) form of fowl pox, with severe involvement of the mucous membranes, or a condition caused by vitamin A deficiency, which is characterized by the formation of white, nonodorous pustules in the mouth, pharynx, and esophagus.

## FOREIGN BODIES IN THE PHARYNX AND ESOPHAGUS

As has been mentioned, cockleburs have been known to lodge in the esophagus of young chickens and turkeys. The base of an electric light bulb has been found in the esophagus of a turkey. It is unusual for birds, because of the anatomical structure of the esophagus, to have foreign bodies lodge in this organ. A specific disease known as vitamin A deficiency or "nutritional roup" sometimes affects the inner surface of the entire organ. This, however,

will be discussed under nutritional diseases. Tumors of this organ have never been observed by us.

## ANOMALIES OF THE CROP (INGLUVIES)

This organ is considered as a diverticulum of the esophagus. Conditions affecting the crop which were, in the past, attributed to purely local irritations are now known to be caused in the majority of cases by parasitic worms (Capillaria), protozoan parasites (Trichomonas), or molds. Pendulous crop and impaction, therefore, aside from the specific conditions already mentioned, are the principal abnormalities observed in birds (Fig. 36.4). Pendulous crop and impactions are probably more frequent in chickens and turkeys than other fowls. Pendulous crop and impaction sometimes both occur in the same individual, and perhaps one may be the cause of the other, although Hinshaw (1936) has shown definitely that pendulous crop tendencies are inherited in about 75 per cent of the cases and that the condition may be controlled by selection



Fig. 36.2. The tongue protruding through a complete fistula of the posterior floor of the oral cavity.

and elimination of birds carrying the factor in their genetic make-up.

Impaction of the crop (referring particularly to turkeys) may occur at any age and is more likely to appear in birds fed an improperly balanced diet. If turkeys are not properly fed and are allowed to run on a litter of wheat straw they often take in enough of the straw to impact the crop, also the proventriculus and gizzard, and a portion of the duodenum may become impacted at the same time, causing an engorgement of all four organs and practically paralyzing the digestive tract. Younger turkeys which run on bluegrass pastures, if not properly fed, are also likely to consume long blades of grass, which form a network in the crop and produce a definite impaction which will finally result in the death of the bird. Cases of impaction have also been observed in adult turkeys when poorly fed or running in pastures. The birds may consume large amounts of persimmon seeds, acorns, twigs, etc., producing a hard impacted mass in the crop. Materials other than those mentioned are likely to cause impaction, especially if the birds are not properly fed. Emphasis, therefore, should be placed upon the importance of feeding a balanced diet for the prevention of impaction.

The treatment for the relief of impactions depends upon the nature of the impaction and the kind of material involved. Ordinarily, flooding the crop with physiological salt solution and the massaging of the outer surface of the organ, in order to soften the material, is indicated. A method which has been used and appears to be very effective in overcoming this impaction, regardless of the nature of the impacted material, is emptying the crop by flushing several times with water. It is well to add a tablespoonful of sodium bicarbonate to each quart of the water used in flushing the crop.

A long nozzle bulb syringe (nozzle 6 to 8 inches long) or dosing syringe equipped with a long nozzle that is used for drenching sheep is satisfactory. The bird should be placed on a level table, on its chest with feet and legs stretched back and its head held slightly lower than the level of the table. The long nozzle syringe or bulb syringe should then be filled with water, the nozzle of which should be passed down the throat of the bird and the water expelled into the crop. This should be repeated until the crop is well distended with the water. The water and crop contents may then be removed by holding the beak open with the index finger of the left hand and with the right-hand fingers extended push backward, upward, and forward on the distended crop, at the same time holding the bird's head downward. In removing the solid material with the water from the crop care should be exercised to keep the large pieces coming from the crop from lodging at the base of the tongue, thereby forcing water into the trachea. This can be prevented by the use of the index finger, the tip of which is placed inside the bird's mouth. It may be necessary to fill and empty the crop several times before all the solid material is removed. After the crop is empty each bird may be given castor oil or a bland oil.

Gauze forceps are very useful if a great amount of grass is present or if the contents are in a very firm compact mass. After partially distending the crop with water, pass the gauze forceps into the crop by way of the esophagus. The large mass may be broken up by external manipulation in conjunction with the forceps.



FIG. 36.3. Horny growth from the lower beak of a twelve-year-old parrot affected with tuberculosis.

Large amounts of grass may be removed with the forceps. Obviously care must be exercised to prevent bruising the mucosa of the crop and when passing the mass of impacted material over the upper larynx.

Pendulous crop is characterized by an abnormal position or pendulous condition of the organ. This diverticulum fills with fluid, sand, gravel, or other heavy material which tends to gravitate down forming a pocket, and by its weight and bulk gradually increases the size of the organ. The material becomes sour because it remains in the crop which has lost its normal power of contraction by means of which food is transferred into the stomach. As has been mentioned, this apparently is an inherited characteristic and can be largely controlled by the elimination of breeders which show this tendency.

A satisfactory treatment for this condition has not yet been devised, though it has been reported that success can be obtained by a surgical operation. The bird is hung up by the legs at a convenient height and the feathers removed over the surface of the crop area. The crop is thoroughly washed out by way of the mouth, as described for impaction, with a syringe. Then an elliptical cut is made in the crop and a sufficient amount of the wall removed to reduce the crop to normal size. The wound is then closed by means of mattress sutures and the diet restricted to milk alone for several days after the operation. One veterinarian has reported that 95 per cent of the birds so operated will make a recovery. In our experience, however, operations similar to the one described have not been successful, and practically all the birds developed a permanent fistula of the crop at the site of the operation. The operation, however, might be attempted by experienced operators and prove successful (see also chapter on Diseases of the Turkey).



FIG. 36.4. An extreme case of pendulous crop in a young chicken.

In addition to the condition mentioned, foreign bodies, such as pieces of bone, glass, or glass rods, may puncture the wall of the crop and cause the formation of a fistula and result in fatality. Occasionally, following the so-called lye treatment for the removal of worms, numerous fistulas may occur where the highly basic material has eroded through the crop and skin. This is rather rare, and the accident is more likely to be observed either in the gizzard or in the intestines.

Impaction of the stomach sometimes occurs in connection with the crop. A primary impaction of the proventriculus may occur, although this condition is usually associated with both the impaction of the crop and of the gizzard. When impaction occurs in the stomach, it is more difficult to treat since it is impossible to remove the impacted material through the mouth as is done in the case of impaction of the crop. Attempts have been made to remove material both from the gizzard and from the stomach by surgery, but the results of these operations are usually fatal. Because of the strong muscular action of these two organs, it is not always possible to close permanently the incision by means of sutures. The administration of oil is recommended with the hope that the impacted material may be loosened and induced to pass down through the gizzard and intestines and thus out of the body. If this treatment does not succeed, the condition usually results in a fatality.

Aside from the *Tetrameres americana,* and other parasites which may affect the proventriculus, this organ is usually free of other serious conditions (Durant and Knight, 1941).

## THE GIZZARD (VENTRICULUS)

The gizzard (muscular stomach), because of its structure, including both the large muscles and tough horny lining membrane, is normally quite resistant to many changes. This organ is able to exert a tremendous pressure per square inch on objects which are taken into the organ for maceration or grinding. Because of this ability to compress, if sharp objects are swallowed by the fowl, it is not uncommon for them to be forced through the walls of the gizzard, sometimes in two directions at once (Fig. 36.5). This results in a chronic local peritonitis, and there is a tendency to produce paralysis of the organ due to the injury, thereby not only causing illness, but preventing the normal passage of food through this organ. Because of the nature of this organ, the chances for repair or remedy in such conditions are poor. The muscular development and the severe pressure brought to bear, make it impossible to remove the foreign bodies and to close the wounds with sutures. Although the authors have attempted this in several cases, the birds succumbed. Exposing the birds to foreign objects that might cause a rupture in the wall of the gizzard should be avoided. According to Joest (1919), the caudal part of the muscularis intermedius is most frequently injured or penetrated.

The pathology caused by the gizzard worm and gizzard erosion, which apparently is caused by a vitamin deficiency, are discussed elsewhere. Erosion of the gizzard has also been observed in chickens when treated for roundworms by means of the lye treatment, which consists of adding a rounding tablespoonful of Lewis lye to one-half gallon of shelled oats and one-half

gallon of whole wheat. This mixture is covered with enough water so that it will not scorch and is boiled slowly for 3 hours, after which it is put out and the birds are allowed all they will eat within 3 hours. In the cases observed by us the birds apparently consumed more of the lye-soaked grain than was intended.

In salt poisoning the gizzard is sometimes involved, although the lesions and changes produced by the salt are not very definite. As was mentioned under impaction of the crop, the entrance into the gizzard and the gizzard itself may be impacted. This impaction may extend into the duodenum for



FIG. 36.5. Proventriculus and gizzard of a chicken showing a 4-penny nail protruding from the gizzard. This nail had been swallowed by the bird and had punctured the wall of the gizzard.

several inches. The only hope of relief from this condition is the administration of oil. Where the impaction is severe and large quantities of dry grass or wheat straw have collected, the prognosis is usually unfavorable.

Tubercles may form on the outer surface of the gizzard in tuberculosis, but this is discussed under that disease. Atrophy of the organ may also occur as is sometimes seen in fowl paralysis.

## ANOMALIES OF THE INTESTINES

In discussing the changes which take place in these organs, it should be remembered that most of the changes which formerly were thought to be primary processes are now known to be manifestations of specific diseases.

Salt poisoning is of fairly common occurrence in poultry, especially chickens and turkeys. Salt poisoning may occur in adult hens and in young growing chicks. The lesions in the intestines are quite similar. Observations have shown that most of the salt poisoning in young chickens and turkeys have occurred when a normal ration was fed. Because of the weight of salt and its crystalline form, there is a tendency for it to settle in any coarsely ground feed, and if birds are fed in feed hoppers with slanting sides which

form a "V" and the hoppers are not emptied regularly, that is, all the food is not removed before fresh feed is put in them, there is a tendency for the salt to settle in the bottoms of these V-shaped troughs. Eventually this results in a high concentration of salt in the feed which will produce salt poisoning when consumed by fowls. When adult birds are involved, poisoning is usually due to birds eating quantities of ice cream salt which had been thrown out after making ice cream. In these cases the birds will fill their crops with the salt and die very quickly as a result of salt poisoning. In the case of young birds exposed to gradual poisoning, there is an accumulation of fluid throughout the body (anasarca). There is also an inflammation of the intestinal wall, characterized by large and small hemorrhages at various places along the digestive tract. The inflammation produced by salt varies considerably; the younger the bird, the more severe the inflammation. In adult birds the inflammation tends to localize more than it does in the young birds. The muscles appear wet and glistening. To correct the feeding fault, the removal of all excess salt from poultry will quickly stop losses from salt poisoning.

Foreign bodies quite often cause perforation of the intestines. These perforations may produce chronic peritonitis or, if they are small, they may be encapsulated by the formation of connective tissue. In that case it is not unusual for a large growth to form at the point of penetration, the center of which may contain a mixture of the material from the intestinal tract and detritus from dead cells, which results in the collection of an offensive mass of material, sealed by this inflammatory wall. Usually when these cases are found they are past the stage where it would be practical to attempt surgical interference, since the birds are so low in vitality from the toxic effects of chronic peritonitis that they are poor surgical subjects. Foreign bodies sometimes cause blocking of the intestinal tract, and the contents of the intestine collect above this obstruction, causing great distention. Below the obstruction the intestine will be abnormally small (Fig. 36.6). This condition may exist for weeks and the birds become severely emaciated. A diagnosis cannot be made except by laparotomy or post-mortem examination of the bird in the last stages of the disease or after it dies. Because of the low individual value of a fowl, this kind of operation would seldom be justified, even though an early diagnosis could be made.

## INTUSSUSCEPTION OF THE INTESTINES

The exact cause of intussusception in fowls is not known. It does not occur very often, and it is thought to be caused by some irritation of the digestive tract at the point where the intussusception begins. Cases have been observed in which 6 inches of the intestine had been folded in. Where this occurs there is usually a marked swelling of the parts involved, and usually necrosis of the tissues themselves takes place because of pressure and circula-

tory disturbance. Birds may show signs of sickness for several days before finally succumbing.

It is not uncommon for fowls which are not fed properly balanced rations to develop a paralysis or condition of the vent which partially obstructs this organ, preventing the normal passage of the feces. The result is a gradual accumulation of waste products in the cloaca and in the large intestine anterior to the vent. The large intestine and cloaca are greatly distended and filled with contents. The bird, in trying to pass this material, will strain and emit painful cries, perhaps being able to pass only small quantities at a time. It is probable that this is caused in most cases by a partial paralysis of the posterior part of the intestinal tract or by mechanical block. However, it is usually not observed where well-balanced rations are fed, leading one to suspect that malnutrition might be more important than other causes except pullorum disease in the chick.



Fig. 36.6. Obstruction of the small intestine of a chicken, showing enlargement of the organ anterior to the foreign body lodged in the lumen.

Vent gleet, which might be classified as a specific disease, is a chronic inflammation affecting the vent, in which there is the formation of a tough, yellowish diphtheritic membrane which may encircle the entire vent and produce a very offensive odor, which is so characteristic that one familiar with this condition will recognize it by the odor upon entering a poultry house where this disease is present, especially if several birds are affected.

Vent gleet is most commonly observed in laying hens, although it is sometimes found in the male. It usually starts with a red, inflammatory appearance of the vent and the formation of a diphtheritic membrane. Some investigators think that the initial lesion or beginning of the disease may be the result of the strain placed upon the organ in laying eggs. This inflammation usually starts in one or both canthae of the vent and gradually spreads dorsally and ventrally. The diphtheritic membrane is gradually formed as the disease progresses until the entire vent is covered with this thick, dirty yellow, tough, diphtheritic membrane. If the membrane is removed by force it leaves a raw bleeding surface.

There are a number of theories as to the cause of this disease, and many organisms have been isolated from the lesions. The specific cause of vent

gleet, however, is unknown. At the Missouri Experiment Station, McDougle has treated the condition quite successfully with a 3 per cent C.P. chromic acid solution. This solution is applied to the affected parts by means of a small cotton swab. The part is gently sponged with this solution every third day until the birds have recovered. Data kept on the treatment showed that some will require as many as eight or ten treatments, but a few severe cases



FIG. 36.7. Above, part of the lower digestive tract of a turkey showing the main intestine. Below, a single elongated cecum with two miniature ceca at its apex.

are not cured by this means. It would appear from observations made that birds which respond to the treatment usually recover very quickly. For that reason it is recommended that after three or four treatments, if marked improvement is not observed, it would probably be better to destroy the individual bird rather than to continue treatment. Vent gleet never occurs as an epidemic in fowls. Usually only a few birds in a flock are affected at a time, and formerly it was thought to be a disease transmitted exclusively by the males. This theory, however, does not appear tenable at this time, although when a case occurs in a flock all birds should be examined frequently. Affected birds should be isolated immediately and treated with 3 per cent chromic acid solution. Any that recover can be returned to the flock without danger to the rest of the birds.

Figure 36.7 shows a condition found in an otherwise normally developed turkey. Only one fully developed cecum was present. It is abnormally long, and from its distal part, two distinct cul-de-sacs were present. (A cecum from a normal bird of comparative weight is only approximately three-fourths as long as the single organ shown in the illustration.)

## ANOMALIES OF THE UROGENITAL ORGANS

Fortunately the common oviparous animals are relatively short lived, as selective breeding has made oviposition most frequent and often difficult. The ordinary chicken in relation to its size lays more eggs than any other bird, thereby placing a great load on the general constitution and reproductive system.

Peculiarly, the avian left ovary is functional, likewise the left oviduct, whereas in practically all mammals both ovaries and oviducts are active. This peculiarity of the oviduct often results in a cystic condition of this part of the reproductive tract, caused by the failure of the right oviduct to develop (Fig. 36.8). Ordinarily this cystic organ, 5 mm. to 2 inches in length, is usually found in the posterior part of the abdominal cavity, one end being attached to the cloaca. Its contents may be clear or turbid (Durant, 1937).

The left oviduct is the site of several peculiarities with relation to the eggs. Two yolks may be released from the ovary, and this large mass passing down the oviduct results in a double-yolked egg which often may be several times the size of a normal egg. Oviposition of such a large egg may result in the rupture of the oviduct or tearing of the vent, thereby leading to cannibalism, as other birds are readily attracted to blood, particularly if the birds are white in color.

Nutrition is sometimes suspected as being a cause since egg size is influenced by several factors such as the age of the bird, number of eggs laid, breed, etc. However, usually only one or two birds in a large



FIG. 36.8. Cystic tumor originating in the ovary of a parakeet. The tumor is protruding from the peritoneal cavity after removal of the skin and the abdominal wall.

flock lay extremely large, or double yolked eggs; therefore, the primary factor is probably hereditary. Such birds should be marketed as the eggs are only curiosities.

Eggs of various shapes and sizes may be laid. Large eggs may sometimes contain a small egg complete with shell and yolk. In some cases specific infec-

tions of the oviduct are responsible. Trematodes sometimes invade the cloaca and oviduct. These parasites, and rarely the common roundworm, may be found in an egg.

Unusual torsion may cause the eggs to be elongated, flattened. ridged, or tubelike in shape. However, apparently normal birds may lay eggs so well defined by a strongly convex ring encircling the egg from side to side that the egg is readily recognized as being produced by a certain bird.

Inflammation of the oviduct may also cause a variety of misshapen eggs or may so occlude the oviduct that the egg passes down far enough to receive a complete shell, yet be forced up the oviduct and into the peritoneal cavity. A laparotómy on a bird clinically called a "penguin posture" resulted in the removal of nine well-formed eggs, and yolks of several more. The patient recovered and was marketed.

### EGG CONCRETIONS

Egg concretions in the oviduct or peritoneal cavity are rather common and present a semi-cooked appearance. Masses of yolk material added to a yolk or yolk material gives the concretion, when incised, the appearance of being built up of concentric rings. These formations may be of various sizes and shapes and may reach a diameter of 3 inches or more. Also several small concretions may be found in either the oviduct or peritoneal cavity. When deposited in the peritoneal cavity, inflammation is produced, causing adjacent structures to thicken and often to assume the characteristic appearance of a tumor.

Eversion of the oviduct seems peculiar to some flocks and in part is apparently an inherent weakness. However, oviposition of an unusually large egg may be responsible for the eversion, also any agent causing prolonged irritation of this organ may cause it to be partially forced out through the vent. Unless the affected bird is promptly removed from the flock, other birds usually eviscerate the unfortunate individual.

Treatment of birds that are egg-bound, or that have a prolapsed oviduct, or that have eggs or an "egg concretion" in the peritoneal cavity, is usually a time-consuming operation. Destruction of the bird is advised unless it is of great value. Recurrence of the condition or conditions mentioned above is common, and it is best to remove these individuals from the flock or breeding pen.

### KIDNEYS

The kidneys of the fowl are peculiar in some respects. Rather than being rounded "free" masses as in mammals, they are large, elongated organs closely adherent to the impressions in the ventral surface of the lumbosacral mass. They are relatively large in relation to the gross weight of the bird.

Although tumors may be found occasionally in the kidneys, the most common noninfectious changes are urinary cysts. These are usually blister-like in appearance, although some may be 5 mm. in diameter. They are filled with a clear, brownish, yellow-tinged fluid.

Ureteroceles, when present, usually occur in the area of that part of the ureter adjacent to the kidney surface. Depending upon the exciting cause, ureterophlegma may precede or accompany the development of the uretero-cele.

## REFERENCES

Asmundson, V. S., and Hinshaw, W. R.: 1938. On the inheritance of pendulous crop in turkeys (*Meleagris gallopavo*). Poultry Sci. 17:276.

Durant, A. J.: 1932. Abnormality in the formation of the ceca of a turkey. Vet. Med. 27:525.

———: 1937. Ovarian cyst in a parakeet. Vet. Med. 32:75.

——— and Knight, D. R.: 1941. *Tetrameres americana* (Cram, 1927) found in eastern cardinal in Missouri. Vet. Med. 36:373.

Hinshaw, W. R., and Asmundson, V. S.: 1936. Observations on pendulous crop in turkeys. Jour. Am. Vet. Med. Assn. 88:154.

Joest, Ernst: 1919. Spezielle pathologische Anatomie der Haustiere. Band. I. Verlag. von Richard Schoetz, Berlin. Pp. 432–34.

# CHAPTER THIRTY-SEVEN

## POULTRY SURGERY

By L. H. SCHWARTE, *Veterinary Research Institute,*
*Iowa State College, Ames, Iowa*

☆  ☆  ☆

Avian surgery was given little or no consideration for many years. The low value of the individual fowl in the farm flock made it economically unsound to resort to individual treatment. In recent years, as the result of the growth and development of the poultry industry to its present large-scale production methods, there has been an increased demand for better and more effective veterinary service to reduce poultry losses to a minimum. The comparatively high individual value of show birds and ornamental and game birds, as well as avian pets, has created considerable demand for individual medication and improved surgical methods. Consequently, substantial progress in avian surgery has been made for practical and experimental purposes.

The avian species make excellent surgical subjects because their sensory nervous systems are apparently not as highly developed as those of the majority of our domesticated animals, and consequently, they suffer less from surgical shock. They also respond to both general and local anesthetics if properly administered. Birds also have a very high resistance to pyogenic infection which simplifies effective surgical technique under field conditions with little danger of post-operative infection.

Avian surgery may be classified in two distinct groups, practical or applied, and experimental. The first group includes those operations necessary to relieve certain conditions which are quite commonly encountered in poultry practice. The second group has been developed for the purpose of studying various problems in physiologic and pathologic research. Experimental surgery is of little practical value. It will be treated briefly together with references for the benefit of those desiring this information. Burrows (1936) developed a technique for the removal of the gizzard of the domestic fowl. Sloan (1936) successfully removed the yolk from newly hatched chicks. Durant (1926), Schlotthauer *et al.* (1933), and Delaplane and Stuart (1933) have developed surgical techniques for cecal abligation in an effort to control infectious enterohepatitis in turkeys. Some of the surgical methods developed in poultry research have contributed much toward our present knowledge of poultry physiology and pathology.

**Anesthesia.** Anesthetics are not generally used in poultry practice. They have been used successfully in experimental investigations which involved major operations. The anatomical structure of the avian respiratory system renders the use of inhalation anesthesia rather unsatisfactory. This type of anesthesia cannot be controlled adequately because of the infiltration of inhaled anesthetic into the air sacs which constitute part of the respiratory system in birds. Frequently, excessive amounts of the anesthetic are absorbed, resulting in respiratory failure, cardiac inhibition, and death. Consequently, ether and chloroform have been replaced by some of the more recently developed anesthetics which can be administered intravenously. Chloral hydrate injected intravenously in doses from 0.2 to 0.4 gm. produces a complete anesthesia lasting from 15 minutes to 1 hour. Hole (1933) reported the minimum lethal dose of chloral to be between 0.4 and 0.6 gm. The intravenous administration of sodium amytol solution (0.1 gm. per cc.) is quite satisfactory as a general anesthetic according to Fretz (1932). Injections of 0.5 to 1.0 cc. of this solution is adequate for birds ranging from 4 to 8 pounds in weight. Warren and Scott (1935) consider sodium pentobarbital (nembutal) the most satisfactory general anesthetic for poultry practice. Intravenous injections of 0.5 to 0.75 cc. produce effective anesthesia for as long as 2 hours. The intravenous use of nembutal for turkeys in doses of 1.1 cc. per 5 pounds of body weight was reported by Durant and McDougle (1935) as an effective anesthetic.

Local anesthetics have a limited use in poultry practice. Schalm (1936) reported the successful use of 2 per cent solution of novocain as a local anesthetic. Butyn in a 2 per cent solution is regarded very satisfactory by Sweebe (1925) for local anesthesia of the eye.

## ABDOMINAL SURGERY

Abdominal surgery is limited to a few operations for conditions which are quite common in poultry practice. Caponizing is probably the most extensive operation performed for the production of birds for a specialized market. The premium received for this class of poultry products makes the operation practical and economically profitable. The removal of eggs in the various stages of development from the abdominal cavity of birds is practiced, especially in cases involving birds which are known to be high producers or those which have high individual values. The removal of excessive quantities of fluids which accumulate in the abdominal cavity is often indicated. Cecal abligation has been given considerable attention in recent years in an attempt to control blackhead in turkeys. At the present time, this operation must be considered only as an experimental procedure. An effective method of sanitation and hygiene instituted in the system of management has proven to be more practical and satisfactory in controlling this disease. Because of

the interest shown in this work, a brief review of some of the methods developed will be presented.

**Caponizing.** The castration or desexing of cockerels is commonly known as caponizing. The optimum stage of development is reached at eight to ten weeks of age when the birds reach about 1½ pounds in weight. At this age there are fewer so-called slips, and the mortality can be kept at a minimum. The water should be withheld for 12 to 18 hours prior to operation. This permits the intestinal contents to be reduced to a minimum, causing the intestines to settle away from the testicles as well as the upper wall of the abdominal cavity. Consequently, when the intercostal incision is made, there is a clearer field of operation in the abdominal cavity and less danger of intestinal puncture. A suitable table, proper confinement, adequate light, and proper instruments are essential for satisfactory results.

The cockerel is placed on the operating table on its left side. The wings are securely held together above the body by a suitable restraining device and fastened to the upper side of the table. The legs are likewise secured together and fully extended, being fastened to the lower edge of the table. This position permits free access to the field of operation (Fig.



Fig. 37.1. Showing field of operation for caponizing.

37.1). Some of the soft feathers are plucked from the region of the hips and ribs. The field of operation is cleansed with a piece of cotton moistened with clean warm water or weak antiseptic solution. With the fingers of the left hand, locate the area midway between the last two ribs (6th and 7th) where the incision is to be made. Draw the skin back over the hip and with it the sartorius or thigh muscle with the left hand. An incision is made about ¾ inch long through the skin and intercostal structures, midway between the

last two ribs slightly below the upper abdominal wall (Fig. 37.2). If the incision is too near the vertebral column or too low, difficulty may be experi‐enced in locating and removing the genital organs. The incision of the vein which runs diagonally across the body from thigh to wing should be avoided.



FIG. 37.2. Incision with insertion of spreaders.

The spreader is placed in the incision, the handle of which should be directed toward the back so as not to interfere with the operator. The peritoneal membrane medial to the inter‐costal structures which forms the abdominal air sac is punc‐tured with a sharp hook or probe exposing the abdominal organs. The gonads which are about the size of a wheat kernel can be seen attached dorsally at the anterior extremity of the kidney. These organs are usually yellowish in color but may vary from white to gray or even black. The lower gonad (left) should be removed first by clamping it securely with forceps and twisting it free from its attachments. The upper one (right) is removed in the same way. The spreaders are then re‐moved and the bird is released from the operating table. The skin and thigh muscle slip back in place as the limbs regain their normal positions serving effectually to close the opening in the abdominal cavity.

The principal danger in this operative procedure is death from internal hemorrhage which takes place within a few minutes following rupture of a primary blood vessel. The testicles are located at a point where the right and left iliac veins converge into the posterior vena cava (Fig. 37.3). The aorta is situated below the vena cava when the bird is on the operating table. The spermatic vessels which supply the circulation of the testicles are ruptured when the testicles are removed; but in the young cockerel these vessels are so small that serious hemorrhages are seldom experienced. The operative field

is such that injury to the major blood vessels can be avoided with ordinary care.

Electrical instruments have been devised and sold on the market for the removal of the gonads to take the place of the more commonly used instru-
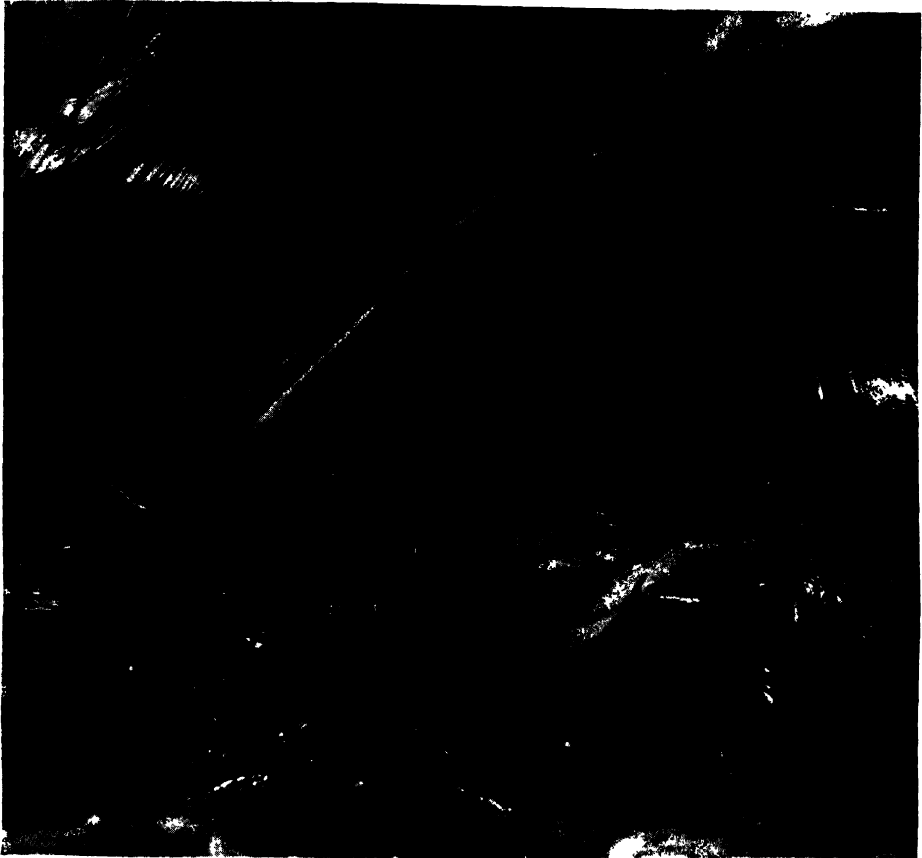


FIG. 37.3. Ventral view showing the relative position of the gonads to that of the major blood vessels in the operative field. 1—left testicle slightly posterior to the right. 2—vena cava formed by the union of the right and left iliacs (3). 4—dorsal aorta. 5—coccygeal vein. 6—mesenteric artery. 7—operative incision on the right side through which both the gonads are removed. (Kansas Agr. Exper. Sta.)

ments. They have the advantage of cauterizing the tissue to which the gonads are attached and possibly reduce the number of slips. They are not so easy to manipulate as some other instruments and not suitable for use under field conditions where electricity is not available.

The removal of the entire organ is very essential in order to prevent so-called slips. Any fragments which remain intact may result in the proliferation of this reproductive tissue, and the operation will fail to achieve the

desired objectives. The secretions from the incised tissues rapidly form hard crusts or scabs. It takes much longer for the healing of the incised tissue to become complete. A certain percentage of operated birds will develop wind puffs which are caused by the air escaping from the body cavity through the intercostal incision becoming enclosed in the subcutaneous tissues. This condition is relieved by puncturing the skin and releasing the enclosed air. The birds should be transferred to clean houses with plenty of clean litter on the floors. They should be confined and kept as quiet as possible for a week or 10 days following caponizing after which they may be given free range and managed to produce the maximum growth and development.

Considerable work has been done in an effort to produce capons by the use of chemicals rather than by the usual operative procedure. Guinn (1944) reported a method which apparently is quite effective and may prove to be of practical importance. Male birds, ranging in age from eight to ten weeks, received a highly compressed pellet containing an average of 15 milligrams of diethylstilbestrol implanted under the skin of the neck. An incision about one-fourth of an inch long was made in the skin of the neck and the pellet was introduced into the incision and pushed about one inch forward from the point of incision. The treated birds gradually lost all male characteristics and to all external appearances and tissue examination could be considered as true capons. There was no recurrence of male characteristics up to the time at which the birds reached five or six pounds in weight. Mature male birds receiving diethylstilbestrol pellets showed considerable improvement in body fat and quality of meat. Lorenz (1945) found that the subcutaneous implantation of diethylstilbestrol was much more effective than the oral administration of this agent. It should be remembered that such subcutaneous implants of any product should be made in the discarded or inedible portions of the bird, such as the upper cervical region. The consumption of the tissue containing either a whole or residual diethylstilbestrol pellet might cause an unfaborable reaction in those who consume such material.

**Cecal abligation.** The surgical technique developed for this operation has been reported by Durant (1926) for fowls and at a later date for turkeys with reference to the control of blackhead (Durant, 1930). The general procedure is similar to that for caponizing. The incision is made on the left side between the last two ribs, as the junction of the ceca and the intestine is located opposite this point. The proximal terminations of the ceca can be easily lifted through the incision to a convenient position for ligation. Each cecum is occluded by means of two catgut ligatures about 4 mm. apart as close as possible to the junction of the ceca and the intestine. These ligatures should be drawn tight enough to close the lumen of the ceca, and yet not tight enough to cut through the cecal wall. The organs are then replaced and the incision is closed by a single suture. Durant reported that if the ligature is passed around the two adjacent ribs and drawn tight enough

effectively to close the margins of the incision, the subsequent danger of wind puffs is practically eliminated. The ceca do not become necrotic because the blood supply to these organs is not destroyed. Atrophy of the ceca from disuse follows in most instances, and they become sealed pouches suspended on the mesenteric ligaments. Occasionally, both ceca may become greatly enlarged in from seven to thirty-two months after the operation. While the complete occlusion of the ceca was effective in preventing blackhead in turkeys, the mortality was so great as to make the procedure economically unprofitable.

The use of aluminum clamps in place of the cecal ligatures as reported by Delaplane and Stuart (1933) reduced the mortality considerably, but this method did not entirely prevent the ceca from resuming their functional activity.

A different operative technique was developed by Schlotthauer and his associates, which proved to be more successful. They made the abdominal incision medial and parallel to the left pubic bone about 1¼ inches in length. A small aneurysm needle was slipped through the mesentery under the ceca near their proximal terminations and the organs exposed through the abdominal incision. A small hemostat was placed across both ceca through the aperture in the mesentery made by the aneurysm needle, and the cecal walls were completely crushed by the hemostats. Silk ligatures were placed above and below the crushing clamp. No losses were reported from this operation, and the resumption of functional activity of the ceca was not observed.

**Egg retention.** The retention of eggs in the posterior portion of the oviduct is not uncommon in fowls during heavy production. This condition may be caused by temporary suspension of the normal physiological activity or may be caused by an obstruction. Abnormally large eggs also may be rather firmly lodged in the oviduct, which require manipulation or surgical removal.

In many cases where there is no indication of pathological conditions, the retained eggs may be removed easily without surgical methods. A lubricant such as mineral oil may be introduced into the oviduct. The forefinger is inserted through the vent; gentle pressure on the abdomen is exerted with the other hand forcing the egg toward the vent. In cases involving eggs of excessive size which cannot be removed without injury to the tissues of the oviduct and cloaca, the egg may be moved posteriorly by manipulation to a position where the shell may be seen through the vent. The shell is then punctured with a sharp instrument after which the egg contents and shell fragments are removed. The cloaca and posterior portion of the oviduct may be irrigated with a cool, mild antiseptic solution to reduce the inflammatory reaction. In cases where it is apparent that tumors are responsible for the obstruction, the fowl should be destroyed.

**Removal of eggs from the abdomen.** The accumulation of eggs in the

abdominal cavity occurs in fowls during periods of high production. The abdomen becomes greatly distended, often reaching the ground (Fig. 37.4 A and B). The feathers are removed from the operative field on the abdomen below the vent. The surface of the skin should be cleansed and disinfected with a mild solution of a suitable antiseptic. General anesthesia may be used effectively. An incision about 3 inches long is made through the skin and abdominal wall between the xiphoid terminal of the sternum and the cloaca. The eggs may be removed from the abdominal cavity by manipulation and pressure on the abdominal wall. The incision is then closed by using a con-
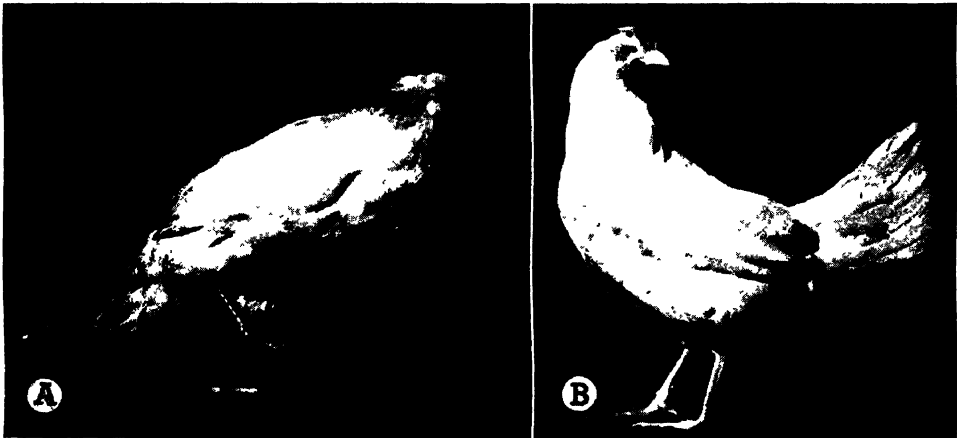


FIG. 37.4. A—characteristic posture of hen with ova in the peritoneal cavity before operation. B—the hen 8 days after operation. (McKenney, Jour. A.V.M.A.)

tinuous catgut suture. McKenney (1929) reports continuous production following a successful operation.

**Ascites.** This condition is defined as an excessive accumulation of serous fluid in the abdominal cavity. The excessive fluid may be removed by inserting a hollow needle or trochar through the skin and muscles of the abdominal wall. This will effectively relieve the internal pressure in the abdomen but will not eliminate the cause. Kaupp (1933) reported the recurrence of this condition in typical cases. This procedure can only be recommended in cases of valuable birds.

### MINOR SURGICAL OPERATIONS

There are numerous surgical operations of a minor nature which are frequently indicated in poultry practice. The majority of these cases respond to ordinary care but if neglected may develop into more serious conditions. Only the more common conditions requiring minor surgical treatment will be discussed.

**Wounds.** The majority of wounds are usually of a minor nature and need

no special care, but more serious injuries require treatment. If the wound is extensive enough to necessitate suturing, the feathers should be removed about the margins of the wound. This area should be thoroughly cleansed with warm water and bathed with a mild antiseptic solution. If the suturing of the muscles or subcutaneous structures are indicated, the loose fragments of the tissue and all necrotic material should be removed. Clean, smooth surfaces of any tissue which are brought into apposition by sutures heal rapidly. Interrupted catgut sutures are preferable for this purpose, as the margins of the wounds are effectively held in place and the sutures are absorbed. It is advisable to employ an independent line of sutures for both muscle tissue and skin. The skin sutures may be either catgut or silk; the former are absorbed and the latter may be removed after healing has taken place. The wound may be dressed with a suitable dusting powder or pulverized boric acid.

**Abscesses.** Abscesses may result from the infection of any bruised or incised tissues. The most common abscess formation encountered in poultry practice is that of the feet which is commonly known as "bumblefoot." Injuries to the feet are subjected to repeated infections by various organisms which contaminate the floors of poultry houses and pens as well as the soil. The infected wounds become enlarged and highly sensitive preceding abscessation. The abscesses must be opened and the accumulated pus should be scraped out manually as avian pus usually is of a cheesy consistency. The abscessed area should be thoroughly cleansed with an antiseptic solution or painted with tincture of iodine. The bird should then be placed in a clean pen. Bandaging of the wound will aid in preventing further infection.

**Foreign bodies.** Foreign bodies are often lodged in the crops of birds. In most instances surgical removal is indicated. The feathers are removed along the medial line in the region of the crop. The surface of the skin should be cleansed and painted with tincture of iodine. Local anesthesia can be used effectively. The bird is held so that the operative field is conveniently accessible. An incision is made along the medial line avoiding the major blood vessels, through the skin and crop wall sufficiently large for the removal of the foreign body. The liquid contents of the crop are absorbed with damp cotton sponges. The foreign body may be removed by manipulation or by use of suitable forceps. The wall of the crop and the skin should be closed by independent lines of sutures. Dusting powder may be applied to the wound. Feed and water should be withheld for about 12 hours after which the bird should be fed sparingly for a few days.

**Crop impaction.** The ingestion of large quantities of bulky or dry feed frequently results in the over-distention of the crop and the inhibition of the normal physiological activity. In some cases a rubber tube may be inserted in the esophagus, and the injection of water into the crop may soften the crop

contents sufficiently to relieve the condition. When the removal of the crop contents is indicated, the operative technique used in the removal of foreign bodies may be successfully employed. In cases of greatly enlarged or pendulous crops, a portion of the wall may be removed before suturing. The postoperative care of the bird is similar to that described in the discussion of foreign bodies.

**Amputation of comb and wattles.** The surgical removal of the comb and wattles is commonly known as "dubbing" and "cropping" in poultry prac-
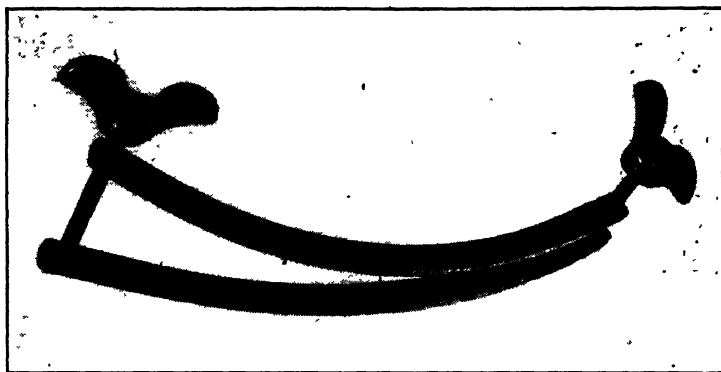


FIG. 37.5. Dubbing clamp. (Schalm, Jour. A.V.M.A.)

tice. Extensive injury, edema, or infection of these appendages indicate removal. The abnormal development of the combs and wattles frequently make it advisable to remove them in order to prevent possible injury. The operative technique developed by Schalm (1936) has been highly satisfactory. The combs of birds are very vascular, and their removal may be followed by profuse and often fatal hemorrhage unless proper operative technique is employed. A suitable local anesthetic such as a 2 per cent solution of novocain in combination with adrenalin or another hemostatic agent can be used effectively. A special clamp for this purpose (Fig. 37.5) is applied to the base of the comb and tightened sufficiently to arrest the circulation of the appendage (Fig. 37.6 A). The comb is amputated with a scalpel or scissors one-eighth of an inch above the clamp following the curvature of the clamp. The cut surface is thoroughly seared with a hot iron and the clamp is removed. The small posterior portion of the comb remaining is removed and seared (Fig. 37.6 B). Complete healing follows as a rule in 30 days.

The wattles may be removed with a heavy pair of shears followed immediately by the application of a suitable astringent such as iron subsulfate. The arterial hemorrhage can be controlled with hemostatic forceps. These operations can be performed at any age, but less hemorrhage is experienced and more rapid healing is observed in younger birds.

**Trimming of claws and spurs.** The trimming of the toenails of poultry

and pets including canaries, parrots, and cage birds can be done easily with heavy surgical shears. The sharp points and edges should be removed with a flat nail file. The spurs of male birds are frequently removed because of injuries inflicted during fights. During the breeding season injuries to the backs of turkey hens caused by the spurs of the males may be quite serious and consequently the removal of the spurs is indicated. A pair of canine bone shears is an ideal instrument for this purpose. These appendages can be amputated with little or no hemorrhage if properly done. In turkeys it is
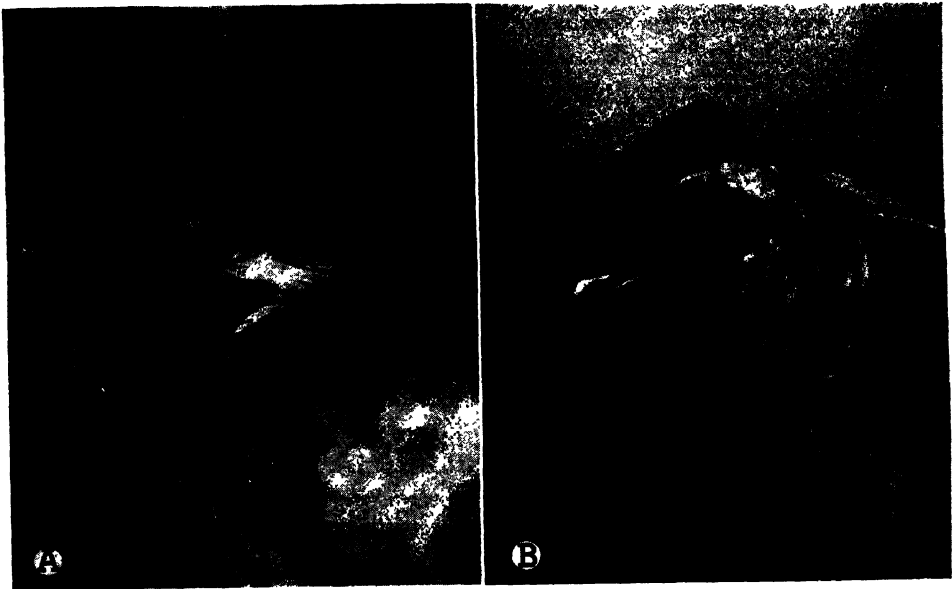


Fig. 37.6. A—cockerel with dubbing clamp applied. B—cockerel 24 hours after being dubbed. (Schalm, Jour. A.V.M.A.)

well to trim the toenails down almost to the corium. If the toenail should be cut too short, tannic acid powder or any other suitable astringent will effectively control the hemorrhage.

**Prevention of spur development.** When large numbers of birds in a breeding flock are kept together it is often advisable to prevent the development of spurs on the male birds. The technique developed by Smith (1932) is simple and effective. This treatment is applied when the spur cap has started to develop and is not over ¼ inch long. Male birds reach this stage of development at ten to sixteen weeks of age, depending on the breed. The spur caps are cut off close to the leg, and a stick of potassium hydroxide is applied and rubbed well into the wound. The cauterizing action of the potassium hydroxide arrests the hemorrhage, destroys the embryonic spur tissue, and prevents the subsequent development of spurs.

**Flight control.** It is often necessary to arrest the flight of birds either

temporarily or permanently. Temporary flight control can be secured by brailing or clipping. Brailing is accomplished by tying one wing closed with a soft cord or bandage. Wing clipping is commonly practiced by poultrymen which consists of cutting the first ten flight feathers close to the wing on one wing only. Clipping is effective in adult birds for a period of about one year while this procedure must be repeated in young birds every month.

The removal of the last segment of the wing at the joint is referred to as pinioning. A tourniquet should be applied to the wing to control hemorrhage. The distal segment of the wing may be removed at the joint with a scalpel or a strong pair of shears. The wound should be thoroughly cauterized with some good caustic agent to prevent hemorrhage and subsequent infection. This operation should be performed on one wing only, and should not be done just prior to or during the breeding season. This operation is best performed in game birds at one week of age.

Permanent flight control may also be obtained effectively by tenectomy which consists in the removal of a section of the tendon which extends along the under side of the wing and runs parallel to the major blood vessels. Pinioning and tendon resection are recommended for game birds bred in captivity.

**Debeaking.** The removal of the tip of the upper segment of the beak is known as debeaking. This operation is indicated to control cannibalism and prevent fighting in male birds. The technique developed by Kennard (1937) has been quite effective. The tip of the beak is not cut off but is separated from the deeper structures by traction or tearing. A short cut is made into one side of the beak only, extending into the margin about 1/16 to 1/8 of an inch (depending on the size of the beak) at a point 1/8 to 1/4 inch posterior to the tip. The flat side of the knife blade is placed against the cut portion of the beak and raised to loosen the edge. The tip is torn off by applying traction toward the opposite side and down toward the lower mandible. This procedure causes little discomfort to the bird and practically no hemorrhage. This removes the tip of the beak rendering the bird harmless for two or three weeks. Debeaking does not prevent the bird from eating or drinking nor does it materially affect the egg production in hens.

**Draining sinuses of the head.** There are several avian diseases associated with infection in the sinuses of the head. In the domesticated fowl the exudate which accumulates in the sinuses rapidly becomes solidified or caseated. In turkeys the exudate remains more or less liquid in form especially in the early stages of sinusitis. The consistency of the exudate can be ascertained by palpation of the enlarged sinuses. If the exudate is solidified or caseated, the surgical removal of the exudate and the irrigation of the sinuses are indicated. The incision of the swollen sinus is made at the anterior ventral margin with a pointed scalpel, making an incision from 3/8 to 1/2 inch in length. The exu-

date may be scraped or curetted out of the sinus and an aqueous solution of silver nitrate applied to the mucous membranes of the sinus. Madsen (1938) reported that a 4 per cent aqueous solution of silver nitrate is slightly more effective than a 2 per cent solution for this purpose. It may be necessary to repeat the application of the silver nitrate solution before the secretion of the exudate is arrested.

The method of treatment recommended by Hinshaw (1937), especially suitable for the treatment of sinusitis in turkeys, consists of the withdrawal of the serous or gelatinous exudate from the sinus with a syringe and hypodermic needle, followed by the injection of silver nitrate solution into the cavities of the sinus. A 5 or 10 cc. syringe fitted with an 18-gauge needle 1½ inches in length is suitable for the aspiration of the exudate from the sinus. The needle should be inserted through the skin and sinus membranes into the sinus. The withdrawal of the syringe plunger removes the fluid exudate from the sinus cavity. About 1 cc. of 4 per cent silver nitrate solution is then injected into the sinus and distributed over the entire surface of the sinus mucosa by gentle massage. Excessive quantities of the silver nitrate solution should be avoided. This method of treatment has been extensively used with satisfactory results. (See sinusitis discussion under turkey diseases.)

**Tumors.** Neoplastic formations are commonly encountered in poultry practice. According to Feldman (1932) birds are subject to many different types of neoplasms involving practically all tissues and organs. From the clinical standpoint, we are interested in two distinct types—benign and malignant. The benign tumors are localized and develop from a central focus. The malignant tumors spread to the surrounding structures and are capable of establishing multiple foci in the course of their development. Because of the common recurrence of malignant tumors, surgical removal is seldom effective.

Benign tumors may attain such a size as to interfere with the normal physiological processes of the bird. When they occur on the exterior surface or in the superficial structures of the body, they can be removed surgically. Local anesthesia may be employed successfully. The neoplastic tissues are excised and the wounds treated as the local conditions may indicate. Internal tumor formations are seldom diagnosed in the living bird. In cases where this condition is suspected, it is advisable to destroy the bird.

**Fractures.** Fractures of the long bones of the wings and legs of birds are not infrequent. Treatment is warranted only in the case of exceptionally valuable birds or in pets. Simple fractures heal readily in a week or 10 days if the fracture is properly reduced and the limb is immobilized. Suitable splints can be fashioned from pieces of light wood, quills, or cork which can be held in place effectively with gauze bandage coated with sodium silicate. In cases of compound fractures, windows may be left in the cast for the pur-

pose of dressing the wound with a powdered antiseptic. Care must be taken not to obstruct the circulation of the limb when applying the splints and bandage.

## REFERENCES

Burrows, W. H.: 1936. The surgical removal of the gizzard from the domestic fowl. Poultry Sci. 15:290.

Delaplane, J. B., and Stuart, H. O.: 1933. Cecal abligation of turkeys by the use of clamps in preventing enterohepatitis (blackhead) infection. Jour. Am. Vet. Med. Assn. 83:238.

Durant, A. J.: 1926. Cecal abligation in fowls. Vet. Med. 21:392.

———: 1930. Blackhead in turkeys—surgical control by cecal abligation. Mo. Agr. Exper. Sta., Res. Bul. 133.

——— and McDougle, H. C.: 1935. Blackhead in turkeys. Mo. Agr. Exper. Sta., Bul. 358:104.

Feldman, W. H.: 1932. Neoplasms of Domesticated Animals. W. B. Saunders Co., Philadelphia.

Fretz, V. C.: 1932. Anesthetizing poultry. Vet. Med. 27:109.

Guinn, A. H.: 1944. Chemical capons. Vet. Jour. 100:241.

Hinshaw, W. R.: 1937. Diseases of turkeys. Calif. Agr. Exper. Sta., Bul. 613.

Hole, N.: 1933 Chloral hydrate as a general anesthetic for the fowl. Jour. Comp. Path. and Therap. 46:47.

Kaupp, B. F.: 1933. Poultry Diseases. 6th Ed. Alexander Eger, Chicago. P. 144.

Kennard, D. C.: 1937. Chicken vices. Bimonthly Bul. No. 184, Ohio Agr. Exper. Sta. 22:33.

Lorenz, F. W.: 1945. The fattening action of orally administered synthetic estrogens as compared with diethylstilbestrol pellet implants. Poultry Sci. 24:91.

Madsen, D. E.: 1938. Sinusitis of turkeys. Utah Agr. Exper. Sta., Bul. 280.

McKenney, F. D.: 1929. Operative relief for ova in the peritoneal cavity of the chicken. Jour. Am. Vet. Med. Assn. 74:1067.

Schalm, O. W.: 1936. Special technics for dubbing and cropping chickens. Jour. Am. Vet. Med. Assn. 89:713.

Schlotthauer, C. F., Essex, H. E., and Mann, F. C.: 1933. Cecal occlusion in the prevention of blackhead (enterohepatitis) in turkeys. Jour. Am. Vet. Med. Assn. 83:218.

Sloan, H. J.: 1936. The operative removal of the yolks from newly hatched chicks. Poultry Sci. 15:23.

Smith, L. W.: 1932. Preventing spur development on male birds. Poultry Sci. 11:241.

Sweebe, E. E.: 1925. Butyn in avian surgery. Vet. Med. 20:491.

Warren, D. C., and Scott, H. M.: 1935. The time factor in egg formation. Poultry Sci. 14:195.

# VICIOUS HABITS AND MISCELLANEOUS CONDITIONS

By L. H. SCHWARTE, *Veterinary Research Institute,*
*Iowa State College, Ames, Iowa*

☆   ☆   ☆

Vicious habits are those undesirable traits which develop within the flock, causing injury or possible death to individual birds and resulting in an economic loss in poultry and poultry products. These vices include cannibalism, flying over fences, egg eating, hiding of eggs, and pica. Vicious habits occur in practically all species of poultry including game birds kept in captivity. These characteristics have developed at times to such proportions in flocks of chickens that they have become of considerable economic importance.

**Cannibalism.** Cannibalism is a vice frequently observed in poultry wherein birds are attacked by their pen-mates, resulting in injury or death. As a rule the bodies of the victims are partially consumed by the offenders. Cannibalism may be manifested in various forms including feather pulling and toe pecking, as well as head, wing, tail, and vent picking. These various forms of cannibalism occur in all breeds of domesticated fowls, but according to Weaver and Bird (1934) the light breeds of the Mediterranean class are much more susceptible to these vices than the heavier breeds of the American and Asiatic classes. Chickens of all ages are subject to these vices. If they are prevented among chicks, these habits are less likely to occur after the birds reach maturity. Some of the causes of these various forms of cannibalism are overcrowding, overheating, confinement in restricted areas with insufficient exercise, and quantitative or qualitative nutritional deficiencies.

According to Kennard (1937) overcrowding and too close confinement are the most frequent errors in poultry management conducive to the development of vicious habits. The minimum requirement recommended for chicks in the brooder house is 0.5 to 0.8 square foot floor space per bird. Growing pullets should have 1 square foot, and birds in production 3.5 to 4 square feet. The floor space per bird can be reduced when the flock is given access to range. Ample space reduces the incidence of disease, parasitic infestation, and the development of vicious habits. The overheating of the brooder house, insufficient ventilation, and excessive light together with

faulty management and unsanitary conditions may be responsible for the discomfort of the birds which may result in the development of vicious habits. The prompt removal of dead or injured birds should not be neglected.

Feather pulling is most frequently observed in flocks kept in close confinement with lack of sufficient exercise. Food and mineral deficiencies may also be contributing factors. The irritation caused by external parasites may induce feather pulling (see external parasites). Hungry chicks are more inclined toward cannibalism than well-fed chicks.

Toe pecking commonly occurs in young chicks, especially to those confined in brooders. Overcrowded brooders which are not properly cleaned and equipped are responsible for the accumulation of feed and fecal material on the feet of the birds. The irritation resulting from the accumulation of these materials on the feet may start toe pecking and subsequently other forms of cannibalism.

Head, wing, tail, and vent picking are the most vicious forms of cannibalism. Injuries to the comb and wattles due to accidental injury or those resulting from fights may start cannibalistic activities. Wing and tail picking frequently follow feather pulling or external injuries. Vent picking in most instances is associated with prolapse of the oviduct. Birds which are in high production may develop highly congested oviducts which not infrequently are everted with the cloaca through the vent. This condition may also develop in birds that become "egg-bound" (failure to pass the egg in the normal manner). If this condition is not discovered promptly and the individual is left in the flock, an outbreak of cannibalism is apt to develop. The other birds in the flock will invariably pick at the inflamed extruded mass until the victim has been seriously injured or killed. This may be the beginning of a series of similar attacks.

The various forms of cannibalism may be prevented in most instances by proper feed and management. Suitable housing and feeding facilities are essential. Adequate quantities of mineral supplements and green feeds together with well-balanced grain rations are conducive to the good health and comfort of the birds. The increase in the fiber content of the grain ration as recommended by Miller and Bearse (1937) apparently reduces the incidence of cannibalism. Kennard and Chamberlin (1936) reported that feather pulling and other vices in confined birds largely disappeared when whole oats were supplemented to the regular grain ration. In some instances salt deficiencies in the diet apparently decrease the normal food consumption and may result in cannibalism and feather picking which may be corrected by the addition of .5 to 2 per cent salt to the ration for 4 or 5 days. The salt level then can be reduced to the optimum level. The elimination of both internal and external parasites also contributes to the comfort of the flock and

reduces the tendency toward vicious habits. Individual birds in the flock which have cannibalistic tendencies may be "debeaked" to prevent such activity (see poultry surgery).

A rather unusual form of cannibalism commonly experienced in young game birds was reported by Bass (1939). The term "nose-picking" has been used to indicate this vicious habit which indicates the nature of this vice. Young quail most frequently develop this habit when crowded together in very small brooder pens. It may start at any time, usually during the period from two to seven weeks of age. It seldom starts after the eighth week, or if it has already started, it stops about that time. One or more of the larger and stronger birds start pecking the upper bills of other birds resulting in injury to the region of the nostrils. The injury, superficial at first, becomes progressively worse as the result of continuous pecking for several days until the bleeding wound invites further injury. The injured bird is unable to eat or drink, becomes weak and is finally trampled to death or suffocated by the other birds. Within a few days a large number of the birds in the pen may become injured in this manner. If nothing is done to stop this habit, most of them are lost. Birds that recover have badly deformed bills and are undesirable for breeding stock. This vice occurs only when the young birds are brooded under artificial conditions. It seldom develops in large pens on the ground in which there is plenty of soil and litter where they may pick and scratch. It was believed that this form of cannibalism was due to a diet deficiency. Various proportions of fish meal, dried meat meal, and milk products were added to the ration which failed to prevent this trouble. The addition of raw meat to the ration is very effective. The birds are given all the raw meat they will eat, and an abundant supply is kept before them at all times. Under these conditions "nose-picking" does not occur, and if already started, it is soon arrested. Ground beef is perhaps the most suitable meat product for this purpose. Frequently it is necessary to withhold the grain ration temporarily in order to compel the birds to eat sufficient raw meat to serve the purpose. The young birds eat ravenously of this meat supplement after they become accustomed to this feeding program.

**Consumption of eggs.** This vice is especially prevalent in the domesticated fowl, often reaching large proportions in a short period of time. The accidental breaking of an egg on the floor of the house or in a nest may start egg eating by individuals in the flock. The production of soft-shelled eggs which, according to Jull (1930), is most prevalent in the spring of the year when production is at its height, is often responsible for the increased incidence of this vicious habit. The lack of adequate nesting facilities, accumulation of large numbers of eggs in nests, and failure to gather eggs at proper intervals may result in egg breakage which in turn encourages the consumption of

eggs. Mineral and protein deficiencies are believed to cause birds to develop this vice. If one bird in a flock becomes addicted to this habit, others soon follow the same procedure.

The control and prevention of this vice may be accomplished by furnishing suitable and adequate nesting facilities and well-balanced rations, and the removal of habitual offenders from the flock. Stafseth (1934) emphasizes the importance of the prompt removal of floor eggs and the necessity of including sufficient lime in the ration to prevent calcium deficiencies.

**Hiding of eggs.** The primitive instincts of wild birds to hide eggs or "steal nests" is often manifested in all breeds of our domesticated poultry. Its occurrence is less frequent with chickens than with ducks, geese, and game birds. In commercial flocks the incidence is rare, but in the average farm flock stolen nests made in barns, fields, hay stacks, straw stacks, or in various out buildings are not uncommon. Inadequate and improperly devised nesting facilities encourage this undesirable habit. Nests which are dirty and infested with lice or mites may cause birds to seek new and more favorable nesting facilities.

Ducks and geese are more inclined to hide their nests. They are often confined to pens during the early part of the production period. These units are provided with suitable nesting facilities and should not be overcrowded. The birds are confined to these pens until they become accustomed to laying in nests provided for that purpose.

Adequate nesting facilities properly maintained and managed so as to provide the maximum comfort and health of the birds will effectively reduce the occurrence of this habit.

**Flight control.** The flight of fowls over fences which confine them to their respective units frequently results in considerable loss by theft, predatory birds, and animals, as well as damage to gardens, crops, and other personal property. It is also inconvenient and time-consuming to have individuals in a flock which require extra attention daily. This vice may be controlled by clipping the first ten flight feathers of one wing close to the margin of the wing. This procedure is effective in mature birds for about one season but must be repeated once a month in younger birds. Brailing may be more satisfactory for flight control in show birds. In the case of valuable birds in which permanent flight control is indicated, pinioning or tenectomy may be effective (see flight control under avian surgery).

**Pica.** A depraved appetite or a craving for unnatural articles of food is known as pica. In an effort to satisfy this abnormal appetite, the birds frequently ingest various indigestible foreign substances such as small sticks, straw, feathers, or even fecal material. The causes of this condition include dietary deficiencies, heavy parasitic infestation, and diseases of the digestive tract.

This condition apparently was quite common years ago according to the early poultry publications. Since the recently developed principles of nutrition have been applied to poultry husbandry and improved methods of feeding and sanitation have been incorporated in the systems of poultry management, the incidence of this condition has become comparatively rare. Occasionally, pica is reported occurring in small farm flocks and in commercial establishments which are not properly managed.

Dietary deficiencies which may be qualitative or quantitative should be corrected (see chapter on nutrition). In cases in which heavy parasitic infestations are apparent, elimination of parasites is indicated (see chapter on ectoparasites).

### REFERENCES

Bass, C. C.: 1939. Control of "nose picking" form of cannibalism in young closely confined quail. Proc. Soc. Exp. Biol. and Med. 40:188.

Jull, M. A.: 1930. Poultry Husbandry. McGraw-Hill Book Co., Inc., New York. P. 72.

Kennard, D. C.: 1937. Chicken vices. Bimonthly Bul. 184. Ohio Agr. Exper. Sta. 22:33–39.

—— and Chamberlin, V. D.: 1936. Oats for chickens. Bimonthly Bul. 181. Ohio Agr. Exper. Sta. 21:95.

Miller, W. M., and Bearse, G. E.: 1937. The cannibalism preventing properties of oats. Poultry Sci. 16:311.

Stafseth, H. J.: 1934. Diseases of adult poultry. Mich. State Coll., Ext. Bul. 54 (Revised).

Weaver, C. H., and Bird, S.: 1931. The nature of cannibalism occurring among adult domestic fowls. Jour. Am. Vet. Med. Assn. 85:623.

## MISCELLANEOUS CONDITIONS

Numerous miscellaneous conditions occasionally encountered in birds are of relatively little importance. Many respiratory disorders, disturbances of the circulatory system, skin conditions, and eye abnormalities are the results of malnutrition, mismanagement, parasites, and toxemias, or they may be secondary to specific infectious diseases.

Inflammatory reactions of the skin may be due to mechanical injury, chemicals, thermal reactions, or certain toxic conditions. Mechanical injuries are usually superficial and of a minor nature, resulting in cuts, wounds, bruises, and lacerations. The majority of these need no special attention, but the more serious injuries should be cared for as designated under surgical treatment. Injury to the skin by chemical compounds is usually due to accident or carelessness which can be avoided by a proper system of management together with the rational use of drugs and chemicals for therapeutic purposes.

Freezing of the comb and wattles may occur in extremely cold weather, especially when housing facilities are inadequate. In extreme cases in which these appendages become necrotic, surgical removal may be indicated. (See chapter on surgery.) In the case of show birds or valuable breeding stock in which individual treatment is indicated, snow or cold water should be ap-

plied to the comb and wattles as soon as possible. The housing conditions should be corrected so as to prevent subsequent recurrence. Surgical removal of the comb and wattles will eliminate danger from freezing when the temperature cannot be controlled.

Fowls in flocks which have inadequate housing facilities and are confined to their houses often get insufficient exercise, being obliged to remain on their roosts a good share of the time. The feathers covering the sternum are worn off, the skin in that region becomes inflamed and thickened and in many instances cornification follows. The sternal crest may also become curved or deformed. This is more prevalent in cases where mineral deficiencies exist especially in the heavier breeds of poultry. These conditions may be avoided by proper management and nutrition.

Birds are well protected by their feather coat from insect bites and skin irritations such as "nettle rash." However, inflammatory skin reactions from those causes may be encountered during the molting periods. These conditions ordinarily need no special care as the inflammation subsides following the dissipation of the irritating agent.

Subcutaneous emphysema is manifested in birds by the accumulation of air in the subcutaneous tissue caused by the escape of air from any part of the respiratory tract. This condition is most frequently encountered in chickens and pigeons. Puncture wounds penetrating the thoracic cavity, air sacs, lungs, trachea, and crop may cause air to escape to the subcutaneous structures. The fractures of the hollow bones which also form part of the respiratory system may also be a contributing factor. Emphysema occurs more often in the anterior part of the body because of the location of the major part of the respiratory tract. It is believed in some cases that air may escape from the epiphyses of the humerus when imperfections exist. The treatment consists of puncturing the vesicle under aseptic conditions and expelling the accumulated air. This procedure may have to be repeated until the defect in the respiratory system responsible for the escape of air has been corrected.

Vesicular dermatitis or "Sod Disease" usually occurs in flocks on range, especially those ranging on prairie sod. Newsom and Feldman (1920) described the disease which affected principally young chicks and occasionally adult birds in eastern Colorado during the months of May, June, and July. Because of the prevalence of the disease in chicks which ranged over unbroken prairie sod, the authors designated it as "Sod Disease." The disease is first manifested by the formation of vesicles between the toes of young chicks. A general inflammatory reaction follows, causing the entire foot tó become greatly enlarged and sensitive. Several days later the blisters rupture after which thick scabs are formed. In many cases the joints become involved, and portións of the toes become necrotic and drop off. In some cases recovery follows in two or three weeks, leaving the feet and toes severely distorted.

The mortality is from 20 to 90 per cent. Newsom and Feldman did not definitely establish the cause of the disease.

Jungherr (1933) reported an outbreak of arthritis in turkeys caused by staphylococci which resembled in some respects cases of vesicular dermatitis in chickens later reported by Hoffman (1939). In the latter outbreak the entire adult population in a flock of 2,600 White Leghorns became affected by this disease. Vesicles were first observed on the comb, wattles, face, feet, and shanks. Later the vesicles ruptured followed by the formation of scabs (Fig. 38.1 A and B). The egg production was reduced to less than 40 per cent during the course of the disease, and the mortality was estimated at 10 per cent. Other cases reported by Hoffman manifested only inflammatory
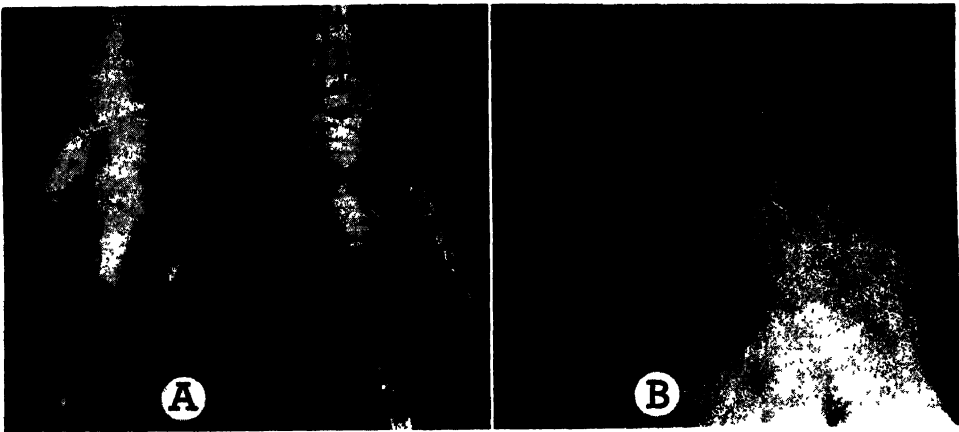


FIG. 38.1. A—scabs on feet and shanks resulting from vesicle formations in the skin. B—scabs on comb as a result of a second attack approximately 5 weeks following the first attack. (Hoffman, Jour. A.V.M.A.)

reactions of the joints of the feet. The organism responsible for this disease was a Gram-positive staphylococcus. Recovery failed to confer an immunity as recurrence of the disease was observed in the same individuals approximately five weeks following the onset of the first clinical symptoms.

Edema of the wattles is manifested by a severe inflammatory reaction in one or both of these appendages followed by an accumulation of edematous fluid. In severe cases the swelling may also involve parts of the head. The exudate may become caseated into hard, cheesy nodules which should be removed surgically. In other cases the inflammatory processes subside, and the edematous exudate is absorbed, leaving a withered appearance to the wattles. The male birds are more commonly affected. The etiologic agent responsible for this condition is not known, but organisms of the Pasteurella group have been isolated frequently from the affected organs and have been suggested as the possible cause.

Inflammation of the uropygial or coccygeal gland may occur in all species of poultry. This sebaceous gland is located dorsally at the base of the tail. It may show a severe inflammatory reaction following the obstruction of the excretory duct. The entire region of the gland may become swollen and decidedly red in color, and is painful on pressure. The exudate becomes caseated unless an incision is made to provide adequate drainage. If the exudate has become solidified it should be removed, and an antiseptic solution such as tincture of iodine should be applied to the wound.

Congenital alopecia is occasionally encountered in newly hatched chicks. Certain avian hybrids develop a partial alopecia which is characteristic of the cross breeding.

Eye disturbances independent of congenital abnormalities are sometimes encountered in large poultry flocks. Mechanical injuries to the eyes are most often experienced by male birds as the result of fights. Foreign bodies occasionally cause injury or inflammation of the cornea and the membranes of the eye. The anatomical structure of the eyes in birds is such that they are less liable to injury than those of mammals. Mechanical injuries to the eye are treated as indicated, the usual procedure is to clean and disinfect the wound to prevent bacterial infection. The eye is an extremely sensitive organ and can be treated easier and more effectively by using a local anesthetic such as butyn or novocain in 2 per cent solution.

Eye complications commonly occur as secondary to specific infectious diseases. Such diseases as leukosis, coryza, diphtheria, and pox frequently present eye lesions. These complications are discussed under the various infectious diseases. Eye disturbances result also from nutritional deficiencies. The avian species sometimes develop tumors of the eye and adjacent membranes. Avian parasites are also responsible for eye disturbances.

The majority of the disturbances of the respiratory system are secondary to specific infectious diseases. The simple cold or nasal catarrh which may under unfavorable conditions develop into pneumonia is especially prevalent in young birds. While the definite cause of this condition is not known, the predisposing factors are believed to include excessive chilling, exposure to rain or dampness, overcrowding, inadequate ventilation, and malnutrition.

Obstruction of the respiratory tract rarely occurs as the result of foreign bodies. Occasionally, some whole grain or foreign object may become lodged in the larynx or trachea. Frequently, it may be retracted through the oral cavity with a suitable instrument. Foreign bodies which cannot be reached in this manner can be removed by performing a tracheotomy. Novocain in 2 to 5 per cent solution is suitable for local anesthesia.

Primary disturbances of the circulatory system are rare and of practically no economic importance. Occasional cases are reported and these are only of academic interest. The vast majority of heart conditions and circulatory disorders are secondary to specific diseases.

Heat prostration in birds may result from excessively high temperatures accompanied by high humidity. The affected birds should be removed to a cool place at once and the body temperature reduced by immersion in cool water. The mortality is high even though prompt and efficient treatment is administered. An ample supply of cool water at all times and properly ventilated houses and nests, together with adequate protection from the sun in hot weather, are effective preventive measures.



Fig. 38.2. Gross bird showing enlarged thyroid. (Kernkamp, Jour. A.V.M.A.)

**Thyroid conditions.** Although a relatively high incidence of goitre in man and other mammals is encountered in certain areas of the United States and other parts of the world, its occurrence among domesticated birds from these areas is rare. Reports on large numbers of autopsies on poultry rarely mention thyroid alterations.

Kernkamp (1925) reported two cases of goitre in 2,409 autopsies on poultry from an area that lies within the so-called goitre belt of the United States. These cases occurred in chickens which were submitted for examination by the same person on succeeding months. The thyroid gland from one of the chickens measured 3.2 × 2.5 cm. (Fig. 38.2). The acini were greatly distended and filled with colloid. The acinal epithelium consisted of a single layer of low flattened epithelial cells. Little or no interacinal connective tissue increase was noted. Both cases were diagnosed as simple colloid goitre.

Balas in 1906 at Budapest reported a case of goitre in a two-year-old cock. Surgical removal of the gland was followed by death. Fox (1923), who autopsied various orders of birds in the Philadelphia Zoological Gardens, found three cases of colloid goitre, four cases of thyroid hyperplasia with colloid, one hyperplasia without colloid, one papillary adenomatoid hyperplasia, and one malignant hyperplasia. Fox states that when carnivora are fed

thyroid gland preparations they do not manifest toxic symptoms until excessive amounts are given, whereas herbivorous varieties are much more sensitive to this feeding. He also observed that meat-eating animals are more sensitive to removal of the thyroid than are the granivora. His data also show that the evidence of thyroid abnormalities is directly related to the carnivorous habits of the species.

Cruickshank (1930) states that the size of the thyroid gland of poultry may be influenced by the type of food, age, species, and seasons. In proportion to body weight the avian thyroid is heavier than that of mammals. More than three times the percentage of iodine is found in·the bird's thyroid than in that of the mammal. The higher temperature and more rapid metabolism seem to indicate that the avian thyroid is relatively more active than that of mammals.

Thyroidectomy in chicks results in retarded growth and feather development; the first molt in thyroidectomized birds occurs later and progresses less rapidly than in the controls. When thyroidectomy is performed on older birds the above manifestations are less marked (Crew, 1926; Lektorsky and Kusmina, 1935; Schwarz, 1931). By means of hyperthyroidization some investigators have produced acceleration of molting and feather growth associated with partial depigmentation of the new growth. Age, sex, and breed appear to be related to the results obtained (Crew and Huxley, 1923; Greenwood and Blyth, 1929; Hutt, 1927; Sainton and Simonnet, 1931; Vilter, 1934; Zawadowsky, 1932).

Wilgus *et al.*, 1941, and other workers have reported that goitre in chickens could be produced by feeding a diet containing 25 per cent soybean oil meal. They attributed the results to the presence of a "goitrogenic factor" in soybean oil meal. Their studies showed that this factor could be partially inactivated by heat; that feeding of iodine protected the animal against the effect of this factor. (See also chapter on nutrition.) The thyroid gland may also be the seat of various neoplasias and lesions concomitant to specific infectious diseases.

## REFERENCES

Crew, F. A. E.:. 1926. A note on a case of peculiar surgical emphysema in the fowl (thyroidectomy). Vet. Jour. 82:430.

———— and Huxley, J. S.: 1923. Effect of thyroid feeding on growth rate, feathering, and egg production. Vet. Jour. 79:343.

Cruickshank, E. M.: 1930. Factors affecting size and iodine content of the thyroid in fowls. Proc. Fourth World's Poultry Cong., p. 237.

Fox, H.: 1923. Disease in Captive Wild Mammals and Birds. J. B. Lippincott Co., Philadelphia. P. 316.

Greenwood, A. W., and Blyth, J. S. S.: 1929. An experimental analysis of the plummage of the Brown Leghorn fowl. Proc. Royal Soc. Edinburgh 49:315 and 335.

Hoffman, H. A.: 1939. Vesicular dermatitis in chickens. Jour. Am. Vet. Med. Assn. 95:329.

Hutt, F. B.: 1927. The effect of feeding thyroid to fowls. Scient. Agr. 7:257.

Jungherr, E.: 1933. Staphylococcal arthritis in turkeys. Jour. Am. Vet. Med. Assn. 82:243.

Kernkamp, H. C. H.: 1925. Goitre in poultry. Jour. Am. Vet. Med. Assn. 67:223.

Lektorsky, I. N., and Kusmina, N. A.: 1935. Die Rolle der Schilddrüse im Prozess der Gefieder-. entwicklung bei den Hühnerkücken. Biol. Zentralbl. 55:16.

Newsom, I. E., and Feldman, W. H.: 1920. Sod disease of chickens. Colo. Agr. Exper. Sta., Bul. 262.

Sainton, P., and Simonnet, H.: 1931. Hyperthyroidisation familiale chez les Gallinacés. Rev. Pathol. Comp. 31:346.

Schwarz, E.: 1931. Pigmentierung, Form und Wachstum der Federn des Haushuhns in Abhängigkeit von der Thyreoideafunktion. Zeitschr. Wiss. Biol. Abt. D. (Roux' Arch. Entwicklungsmech der Organismen) 123:1.

Vilter, V.: 1934. Action de l'hormone thyroïdienne sur la pigmentation des oiseaux. Compt. rend. Soc. de biol. 117:330.

Wilgus, Jr., H. S., Gassner, F. X., Patton, A. R., and Gustavson, R. G.: 1941. The goitrogenicity of soybeans. Jour. Nutr. 22:43.

Zawadowsky, B.: 1932. Hormone und das Gefieder der Vögel. Endokrinologie 10:23.

# CHAPTER THIRTY-NINE

## POISONS AND TOXINS

By L. H. SCHWARTE, *Veterinary Research Institute,*
*Iowa State College, Ames, Iowa*

☆　☆　☆

The literature contains many reports of acute and chronic poisoning of birds due to the ingestion of toxic substances, but the losses from these conditions are insignificant compared with the losses experienced from various other diseases. The majority of cases of poisoning are accidental or due to a poor system of management. When birds are confined to small units or the supply of natural food on the range is limited due to unfavorable weather conditions, they may consume any succulent food available regardless of its palatability or toxicity.

Losses in birds may be attributed to autointoxication, bacterial intoxication, and poisoning by drugs and chemicals, as well as by various phytotoxins, insects, and food constituents. Some of these agents are comparatively rare, and little is known about them, but the more common ones have been investigated, and the toxic as well as the lethal dosages have been determined. In cases of poisoning in birds, positive diagnoses in most instances are made too late for effective treatment, but if the cause is definitely established, it may be removed and further losses avoided.

The body defenses which act to destroy the toxic action of poisons vary considerably in different animals. There is also a variation in the tolerance of certain species for toxic agents. As the result of extensive investigations, Sherwin and Crowdle (1922) found that the action in the bodies of fowls was similar to that of other animals regarding the detoxication of various poisonous substances.

### AUTOINTOXICATION

Autointoxication may be defined as self-poisoning due to the absorption of the waste products of metabolism or of the products of decomposition within the intestine. In young chicks which are raised under artificial conditions, autointoxication is more frequently experienced as the results of injudicious feeding practices. The feeding of bulky foods rich in crude fiber which may occlude the digestive tract may prevent proper elimination and cause the absorption of decomposed contents of the intestines. Chicks which

are raised in confinement and are supplied with chopped green feed often consume quantities of coarse fibrous stems which obstruct the intestinal tract. This condition occurs most frequently in young poults which are raised in confinement and supplied with green feed containing short pieces of fibrous stems. Large numbers of poults may be lost in a short time following such feeding procedures. The use of hay chaff for litter in poultry houses frequently results in the ingestion of indigestible fibrous material which may cause obstruction of the intestinal tract.

The symptoms observed in birds suffering from autointoxication include loss of appetite, increased water consumption, and depression followed by weakness and prostration. Nervous symptoms typical of a generalized toxemia may appear shortly before death.

Occasionally sudden death occurs among apparently healthy turkeys caused by the consumption of large numbers of grasshoppers without any appreciable amount of other food being taken at the same time. Death is caused by the hard parts of the grasshopper, particularly the spined legs which irritate the mucosa of the digestive tract and frequently puncture the walls of the crop and intestines. Nongallinaceous birds are apparently not affected in this manner.

## BACTERIAL TOXINS

Although the losses in birds attributed to bacterial toxins are not considered to be of great economic importance, they occasionally result in heavy losses in individual flocks. The only organism of this type which is important in the consideration of poultry diseases is *Clostridium botulinum*. No significant lesions are found in botulism, and a positive diagnosis is based upon demonstration of the organism and its toxin.

## MOLDS AND FUNGI

Molds and fungi frequently attack grains and forage crops in the field and in storage when conditions are favorable for the development of these organisms. They frequently produce toxins which are poisonous to mammals and birds, and in some instances have caused considerable losses. As a rule, birds are less susceptible to poisoning by molds or fungi than are the common species of domesticated animals. Moldy grains have long been considered as dangerous for stock feed, but they are invariably fed until losses occur. The appearance of the grain is no index as to its toxicity. Some of the worst-looking grains may prove to be nontoxic while brighter and better-appearing grains may be extremely poisonous. Chickens have been fed moldy corn infected with species of Diplodia, Aspergillus, Mucor or Rhizopus, Penicillium, and various bacterial organisms without any unfavorable results. Scabby barley heavily infested with *Gibberella saubinetii* has been fed to

fowls without apparently affecting their health or egg production. Wheat damaged by the so-called stinking smut was used quite successfully as a poultry feed, although it is generally admitted that the feeding value of such grain is impaired by the action of the smut.

Ergot poisoning, however, is occasionally responsible for extensive losses in poultry. This fungus is associated with rye and may cause toxic reactions in flocks which are fed considerable quantities of rye in their rations. In European countries where rye is employed rather extensively as a poultry feed, ergotism is frequently encountered. The clinical symptoms manifested include loss of appetite, abnormal thirst, diarrhea, vomiting, general debilitation followed by convulsions, paralysis, and death. The comb, wattles, and body extremities may become discolored and necrotic in the less acute cases. Various degrees of gastroenteritis together with degenerative changes in the heart, liver, and kidneys, as well as necrosis or gangrene of the appendages and extremities, constitute the major pathologic lesions found in ergot poisoning of birds.

Even though the practice of feeding salvage grains as well as those infected with molds and fungi is quite common, extreme care should be taken in the selection of the grain constituents, and only feed of good quality should be used as poultry feed.

### DRUGS AND CHEMICALS

The poisoning of poultry by drugs and chemicals is most frequently due to accident, carelessness, or the injudicious use of these products as medicinal agents.

**Ammonium chloride.** This chemical, while not commonly used as a medicinal agent, sometimes has been administered in an effort to prevent ascites or the accumulation of fluids in the abdominal cavity of birds. The nontoxic dose reported by Gallagher (1919) was 15 to 45 grains; the lethal dose was found to be 60 grains.

The clinical symptoms manifested in ammonium chloride poisoning are loss of appetite, depression, progressive weakness, coma, and death. No characteristic lesions can be demonstrated.

**Arsenic.** Arsenical preparations are extensively used in the control and extermination of rodents and insects. When considerable amounts of these arsenical compounds are ingested by birds, toxic reactions may result which frequently terminate fatally. Van Zyl (1929) states that birds are more resistant to arsenic poisoning than horses, sheep, and cattle. Gallagher (1919) considered 5 grains of arsenous acid as the minimum lethal dose. Barber and Hubster (1933) reported that small doses of arsenic fed to birds usually cause slight depression which soon disappears. Such birds develop a marked tolerance to this poison, and fatal terminations occur only following administra-

tion of heavy doses. They considered the lethal dose of arsenous acid to be 4 grains or more at a single ingestion.

The possibilities of birds being poisoned by orchard sprays and grasshopper bait are very remote. Whitehead (1934) claimed that birds will not be injured through picking up well-scattered poisoned bran. Furthermore, poisoned grasshoppers which were killed by feeding on poisoned bran and fed to poultry apparently produced no toxic effect. Cooley and associates (1923) concluded that there is no danger of poisoning birds by poisoned bran if the bait is properly used. Wilson and Holmes (1936) found that chickens will not eat enough poisoned bran or bait (arsenic trioxide) to injure them. They also declared that there is no danger from using the normally edible parts of any chicken fed considerable amounts of arsenic trioxide over a period of three months. Hinshaw reported that arsenic trioxide in dosages of 0.25 to 0.5 gram was toxic for turkeys at eight weeks of age.

The clinical symptoms manifested in birds poisoned by arsenical preparations include drooping of wings and ruffled feathers; spasmodic jerking of the neck which finally becomes twisted to one side; depraved appetite; vomiting of fetid serous fluid; cyanotic comb and wattles; loss of equilibrium; and lowered temperature with little effect on the appetite of the bird.

Small doses of arsenic produce practically no lesions in the digestive tract. Large doses produce a severe inflammatory reaction in the crop, gizzard, and intestinal tract accompanied by a catarrhal exudate. The liver becomes very friable and appears yellowish brown in color. The gall bladder is usually distended. The kidneys are enlarged and frequently undergo severe degenerative changes. The fat becomes soft and edematous and appears orange in color. In chronic cases the heart becomes enlarged and flabby. The blood is scarlet in color and watery, having little if any tendency to coagulate.

**Mercurial poisoning.** Metallic mercury is not poisonous, as it is not absorbed readily in the intestinal tract unless ingested in a very finely divided form, according to Lander (1926a). The oxides are poisonous, chiefly the yellow which is a constituent of paint. The sulfides are not very toxic because of their insoluble properties. The chlorides of mercury which are used as disinfectants and medicinal agents in veterinary and poultry practice are occasionally responsible for toxic reactions in birds. Mercurial chloride (calomel) is tolerated by most species of birds, but geese seem to be particularly susceptible to this compound. Bichloride of mercury (corrosive sublimate) is sometimes consumed in sufficient amounts to produce toxic effects.

The toxic and lethal dose of bichloride of mercury was reported by Gallagher (1919) to be 4 grains for fowls. He found 3 grains to be nontoxic with apparently no unfavorable reaction. Hinshaw points out the danger of using 1:2,000 dilution of bichloride of mercury in drinking water for young turkeys. (See chapter on Diseases of the Turkey.) The symptoms observed in

mercury poisoning include progressive incoordination and leg weakness followed by complete loss of locomotion. The birds may or may not show marked depression. In chronic cases the symptoms are less severe, and the progressive developments are retarded. According to Nunn (1907), when mercury enters the circulation it is not directly eliminated from the body, as it is deposited in most of the tissues, chiefly the liver and kidneys. The lesions found in birds as the result of mercurial poisoning resemble those characteristic of a generalized toxemia. Various degrees of gastroenteritis may be observed throughout the digestive tract with or without distinct hemorrhagic



Fig. 39.1. Kidney of goose showing degenerative changes in region of glomerulus. ×320.

areas. Considerable amounts of greenish gelatinous exudate in the alimentary canal may be sufficient to color the ingested food material. The mucous membranes frequently become necrotic and exfoliate. The kidneys are usually pale in color, showing degenerative changes and often are studded with minute white foci. The liver shows evidence of fatty degeneration. The abdominal cavity frequently contains a thick viscid fluid greenish in color.

Geese seem to be quite susceptible to poisoning with calomel. A single dose of 2 grains or more may cause death in less than 24 hours. The symptoms and lesions produced in geese are similar to those found in other birds. However, the degenerative changes found in the heart, liver, and kidneys are much more severe and may account for the rapid and fatal reaction of calomel in geese. Figure 39.1 shows the extensive degenerative changes in the region of the glomeruli of the kidney and Figure 39.2 shows the formation of crystals

in a necrotic focus of the kidney. These changes were observed in the kidneys of experimental geese which were given 2-grain doses of calomel.

**Boric acid.** Boric acid poisoning is very rare in birds. The practice of using chemicals in the preservation of canned foods in order to inhibit the growth of putrefactive organisms resulted in the addition of boric acid for this purpose. Gallagher (1924) reported that canned string beans to which boric acid was added at the rate of 9 grams per quart were toxic to chickens. The clinical symptoms described were loss of appetite, diarrhea, depression, and progressive weakness followed by coma and death. The lesions produced



FIG. 39.2. Calomel poisoning in goose showing crystals in necrotic focus of kidney. ×320.

were severe gastroenteritis, the mucosa of the crop becoming thickened, necrotic, or gangrenous. Degenerative changes in the kidneys were quite extensive.

**Copper poisoning.** Copper sulfate and Bordeaux mixture are perhaps the most common copper compounds used in agriculture, the former as a medicinal agent and the latter as an orchard spray. In poultry practice copper sulfate, also known as bluestone or blue vitriol, has occasionally been recommended for the medication of drinking water. If sufficient amounts of this chemical are ingested, fatal intoxication is frequently observed. Gallagher (1919) found that 20 grains of the crystalline salt or 15 grains in solution was the toxic and lethal dose for the fowl.

Pullar (1940a) reported the minimum lethal dose in grams per kilogram body weight to be 0.9 copper sulfate crystals, 0.3 to 0.5 copper sulfate when

mixed with twice its weight in sodium chloride, and copper carbonate 0.9. Both copper sulfate and copper carbonate in quantities of 1.0 to 1.5 grams per kilogram body weight were considered the minimum lethal dose for pigeons, while that for several species of ducks ranged from 0.4 to 0.9 grams. Pullar (1940b) considered the maximum daily intake of copper carbonate tolerated by birds to be 0.06 gm. per kilogram live weight for fowls and 0.029 for domesticated mallard ducks. No toxic effects were noted from copper sulfate 1 to 4,000 in drinking water for fowls or domesticated mallard ducks.

According to Lander (1926a) the salts of copper in the stomach form albuminates which are quite soluble and rapidly absorbed. They are conveyed to the various tissues by the blood stream and deposited chiefly in the liver, lungs, and kidneys. The elimination of these products in the bile and urine is rather slow. Clinical symptoms of copper poisoning in fowls depend largely on the amount of the toxic agent absorbed. In mild cases a slight depression may be observed followed by recovery. In fatal cases, a primary stimulation and activity may be noted followed by severe depression and weakness. Coma, convulsions, and paralysis may occur before death. The lesions consist of catarrhal gastroenteritis accompanied by the secretion of a greenish seromucous exudate. Coagulation necrosis of the mucous membranes of the lower esophagus and crop are often observed. Hemorrhages are frequently found in the mucosa of the intestines. Degenerative changes in the liver and kidneys may be quite severe.

**Cyanides.** Hydrocyanic acid poisoning may arise not only through the use of the acid and its salts, but through the consumption of certain species of plants which under certain circumstances generate sufficient hydrocyanic acid to cause fatal cases of toxemia. The highly toxic properties of these compounds make them quite effective in the extermination of rodents, birds, and plant parasites.

Birds are seldom poisoned by cyanides except through accident or carelessness. As a rule, they do not select cyanogenic plants for food except in rare cases when there is no other green feed available. Calcium cyanide is extremely toxic for birds. This compound is used extensively to destroy large numbers of undesirable birds such as sparrows or starlings. This preparation is distributed by dusting machines at night while birds are at roost. The inhalation of the calcium cyanide results in the destruction of the birds in a very short time. Winchell (1925) reported the successful use of calcium cyanide in the destruction of large numbers of domesticated fowls condemned as control measures in an outbreak of European fowl pest. One pound of this compound was sufficient to destroy 2,000 birds within 1 or 2 minutes.

Gallagher (1919) found the toxic dose of potassium cyanide to be 1/10 to 1/2 grain, while the lethal dose was considered to be from 1 to 2

grains. The symptoms develop soon after the consumption of the toxic substance. The bird usually loses its sense of balance, drops to the floor in a comatose condition, and dies in a very short time. The action of cyanides in large doses is so rapid that characteristic lesions are not well developed. The comb and wattles appear cyanotic; the internal organs become congested; the blood is dark and has an oily appearance; bubbles of gas may be seen in the cavities of the heart. Characteristic odors resembling those of bitter almonds can be detected in the blood and congested organs.

**Lead.** The common commercial preparations of lead which may be responsible for lead poisoning include the oxides and carbonates of lead, lead acetate, lead arsenate, and metallic lead. The oxides and carbonates are used in paint preparations. Lead acetate is used in commercial and medicinal products. Lead arsenate is incorporated in orchard and garden sprays, while metallic lead especially in the form of shot has been responsible for lead poisoning in fowls and game birds. Most of the lead compounds are comparatively insoluble in water, but may become more soluble in acid or alkaline solutions. Lead salts in contact with digestive fluids may form albuminates and other more soluble compounds which are readily absorbed and distributed throughout the tissues by the blood stream. Lead compounds may be deposited in various amounts in the liver, kidneys, bones, and nerve and muscle tissue. Elimination of lead from the tissues is slow, and effected by way of the bile, urine, salivary, mucous, and cutaneous secretions.

Lead poisoning in domestic fowls is probably most frequently brought about by the consumption of paint skins. In the case of waterfowl and game birds, lead poisoning has been reported from the ingestion of lead shot which accumulate in areas which are used as hunting grounds. This form of poisoning varies considerably in birds, from a severe fatal form to a mild or chronic type, depending on the quantity and nature of the chemical compound. The clinical symptoms may be mild, followed by recovery or muscular paralysis; depression and emaciation may develop, terminating in death. Constipation is followed by diarrhea, during which a thin, watery, greenish fecal excrement is discharged. The lesions associated with lead poisoning in birds include various degrees of gastroenteritis, depending on the amount and nature of the compound ingested. Degenerative changes in the liver and kidneys are usually quite severe. The stippling which occurs in red blood cells in cases of lead poisoning has been long considered one of the characteristic pathologic changes. Johns (1934) considered the stippling of blood cells as characteristic of a dying cell, and that the selective affinity of lead salts for immature red blood cells caused their early destruction.

Investigations have been carried on in the interests of the conservation of wild life, as to the cause of extensive losses occurring in wild waterfowl. Some of these losses were found to be due to lead poisoning resulting from

the ingestion of shot pellets. Wetmore (1922) reported that the usual number of shot found in the dead waterfowl was 15 to 40. The maximum number recorded was 150, and the average was calculated to have been 25.

Variations in lead poisoning in pigeons were reported by Hanzlik and Presho (1923). They observed a prompt loss in body weight accompanied by progressive paralysis. Clinical symptoms appeared about 8 to 10 days after the introduction of lead shot directly into the crop. They considered the minimum lethal dose to be 0.16 gram per kilogram body weight of metallic lead.

**Naphthalene.** Naphthalene formerly was frequently used in the form of moth balls as a protection against lice and mites in nests. This preparation is fairly volatile and gradually decreases in volume on exposure to air. When the moth balls become small they may be readily ingested by fowls. The loss of 40 fowls in a flock of 400, caused by naphthalene poisoning, was reported by Hudson (1936). The clinical symptoms associated with this form of poisoning include congestion of the comb and wattles, abnormally bright eyes, greenish-black diarrheal excrement, progressive paralysis, and death. Death usually occurs within 3 days following the first appearance of diarrhea. The lesions consist of severe catarrhal gastroenteritis with necrotic areas in the mucous membrane of the crop. The liver is congested and greatly enlarged with numerous small necrotic foci. The strong characteristic odor of naphthalene can be detected in the contents of the crop and gizzard.

**Nicotine sulfate.** Nicotine, the highly toxic alkaloid of tobacco, has been used for many years by nurserymen and gardeners for the control of insects. According to Carpenter (1931) the development of the commercial "Black Leaf 40," a standardized 40 per cent solution of nicotine sulfate, made it possible to standardize the dosage and obtain more efficient results in the treatment of internal parasites of poultry. Apparently mature fowls tolerate greater doses of nicotine sulfate than do mammals or other animals. Various commercial preparations containing nicotine sulfate and other constituents including kamala have been used with varying degress of success in controlling intestinal parasites. Bleecker and Smith (1933b) reported toxic reactions in birds treated internally with "Black Leaf 40." They (Bleecker and Smith, 1933a) also reported the toxic dose of this preparation to be from 0.5 to 1.0 cc. In some cases birds receiving a toxic dose became depressed and prostrated, and died in a short time. Parker (1929) found that baby chicks were quite susceptible to poisoning with nicotine sulfate. Doses of 0.2 cc. in various concentrations were given with the following results: an 8 per cent solution killed all treated chicks; 6 per cent solution was fatal to 70 per cent; 4 per cent solution resulted in the loss of 50 per cent; and 3 per cent solution, though producing a toxic reaction and coma for about 15 minutes, resulted in very few deaths.

The application of nicotine sulfate in the form of "Black Leaf 40" on the roosts of the hen house shortly before fowls go to roost has been quite effective in controlling external parasites. According to Carpenter (1931) nicotine is highly volatile at 100 to 105 degrees Fahrenheit, and is volatilized by the body temperature of the fowl. Cases of severe intoxication have been reported from the improper use of this product. Proper ventilation of the poultry house prevents the possible accumulation of vapors sufficient to produce toxic reactions.



FIG. 39.3. Nicotine sulfate poisoning in chickens showing dilatation of the pupil.

The symptoms observed in nicotine poisoning include severe depression, retarded respiration, cyanosis, and coma followed by death. The lesions usually observed are as follows: congestion of the lungs and liver, ecchymoses of the lungs and heart, congestion of the nictitating membranes, dilatation of the pupil (Fig. 39.3) and a dark cyanotic condition of the blood.

**Phosphorus.** The compounds of phosphorus are seldom responsible for poisoning in birds, but occasionally cases are reported resulting from the ingestion of products containing phosphorus. The two most common forms of phosphorus are the yellow and the red. The former is very active, oxidizing readily, being highly inflammable and extremely toxic. It is incorporated in suitable vehicles as pastes, to poison baits for mice, rats, and other rodents. It is also used in the manufacture of matches, firecrackers, and fireworks. The red phosphorus, which is an amorphous form, is believed to be nontoxic. Occasionally, birds will accidentally consume sufficient amounts of poison baits to produce a fatal toxemia. Domesticated birds on unrestricted range, especially turkeys, are inclined to consumed fragments of firecrackers and fireworks which remain following certain holiday celebrations and pyro-

technic displays. These fragments frequently contain sufficient active phosphorus to cause poisoning. Dissolved or finely divided phosphorus may be absorbed as such, absorption being facilitated by the emulsifying action of the bile and digestive secretions. In the alimentary tract, phosphorus has an extremely irritating and caustic effect. The absorbed phosphorus is deposited in the various tissues through the circulatory system, being eliminated principally through the lungs and urine.

The toxic dose for fowls, according to Lander (1926b), is 0.3 grain. The clinical symptoms observed in phosphorous poisoning include depression, loss of appetite, abnormal thirst, diarrhea, and incoordination followed by progressive paralysis, coma, and death. In very acute cases involving large amounts of toxic material, sudden death may occur without noticeable clinical symptoms. The lesions in peracute cases may not be well developed, although fragments of the toxic material may be found in the digestive tract. Beaudette *et al.* (1933) reported that the characteristic odor of phosphorus can usually be detected in the crop and gizzard. In the less acute cases, varying degrees of gastroenteritis may be encountered. Frequently, areas of necrosis occur on the mucous membranes of the crop, proventriculus, gizzard, and intestines. The liver becomes congested and enlarged, friable, and light in color, indicating extensive degenerative changes. The kidneys also show evidence of severe degeneration. Hemorrhage may or may not appear in the membranes and tissues of various organs.

**Zinc phosphide.** Zinc phosphide ($Zn_3P_2$) was used considerably as a substitute for red squill as a rodenticide. Bait was prepared containing about 5 per cent of this compound which proved toxic for domesticated birds and animals as well as rodents. Hare and Orr (1945) recorded cases of poisoning in both geese and fowls due to zinc phosphide.

Clinical symptoms usually appear in 45 to 60 minutes after the administration of a large dose of this compound. Severe depression and ruffled plumage are the first clinical symptoms observed. Diarrhea and progressive weakness develop later. Birds stand with head hanging under body. The birds gradually assume sternal recumbency, lie on side and usually pass through a period of feeble convulsive movements with head drawn over back and legs extended. A few birds will develop severe convulsive spasms prior to death. Large doses frequently render the bird moribund without any nervous reactions. Sublethal doses are followed by depression and a rather mild digestive disturbance with the passage of greenish feces and in some instances diarrhea

Lethal doses of zinc phosphide for fowls is considered to be between 7 and 15 mg. per kilogram live weight. The repeated ingestion of sublethal doses may result in chronic toxemia.

Post-mortem examination reveals various degrees of congestion with the

accumulation of some serous fluid in the pericardial sac as well as in the abdominal cavity in some cases. Enteritis is usually limited to the upper part of the small intestine. A characteristic pungent odor of phosphorus can be detected in the contents of the crop and gizzard especially in birds having ingested large doses.

**Nitrates.** The nitrates of potassium and sodium have been known to produce poisoning in poultry, the general character of which is similar to that of sodium chloride poisoning. Sodium nitrate is commonly used in the form of Chili saltpetre as a fertilizer and may be mistaken for sodium or magnesium sulfate. Such errors have occured in attempted medication of fowls with fatal results. Guberlet (1922) reported the lethal dose to be from 60 to 70 grains for the average fowl, smaller doses causing digestive disturbances accompanied by diarrhea.

The clinical symptoms most frequently observed are excessive thirst, anorexia, vomition, diarrhea, retarded heart action, subnormal temperature, and cyanotic appearance of comb, wattles, and skin. Muscular weakness develops into progressive paralysis followed by coma and death. In some cases convulsions appear shortly before death. The lesions include varying degrees of gastroenteritis, frequently of a hemorrhagic nature. Degenerative changes may be observed in the heart, liver, and kidneys. In peracute cases the lesions are less distinct.

**Potassium permanganate.** This chemical compound is frequently used as an antiseptic in drinking water for poultry, but is decidedly toxic if administered in greater amounts than recommended for therapeutic use. Mature fowls are apparently not injured by consuming a 1 to 500 solution of potassium permanganate as drinking water for several weeks. Gallagher (1919) reported the toxic dose to be 30 grains. He also reported that an experimental fowl died in less than 24 hours following the administration of the toxic dose. No clinical symptoms were observed prior to death. The lesions consisted of a severe cauterization of the crop wall. The submucosa and skin on the lower surface of the crop were blackened. Extensive blood clots were found in the crop where the tissue came in contact with the chemical crystals. All the other organs were normal. The potassium permanganate apparently did not leave the crop, the caustic action of the chemical compound being localized in the tissues with which it came in contact.

**Sodium bicarbonate.** Sodium bicarbonate is not generally regarded as toxic to man or animals, but apparently has caused toxic reactions in young chickens. Similar reactions were quite frequently experienced when excessive amounts of this agent were used as a laxative. Witter (1936) reported toxic reactions in young chickens which were given a 0.6 per cent solution, and fatal results occurred in birds under eight weeks of age following the administration of a 1.2 per cent solution. The consumption of a 2.4 per cent solution produced toxic symptoms in mature fowls.

The clinical symptoms include depression, loss of appetite, and symptoms of a generalized toxemia. The lesions resemble those found in cases of uremic poisoning in birds. The kidneys are pale in color, greatly enlarged, and engorged with urates. The liver also shows evidence of degenerative changes. The heart may or may not be enlarged, but is generally light in color and somewhat flabby.

**Sodium chloride.** Most species of domesticated fowls are susceptible to poisoning by sodium chloride. Ice cream salt, freezing brine, or fish brine, when accessible to fowls, is often consumed in sufficient quantities to poison them. Gallagher (1919) reported 2½ drams of this salt to be the toxic dose for chickens.

The clinical symptoms include loss of appetite, severe depression, and progressive paralysis followed by respiratory failure and death. The lesions are usually confined to a severe congestion of the mucosa of the anterior portion of the digestive tract. Birds which consume a quantity of rock salt or large salt crystals frequently develop a severe enteritis. Krakower and Goettsch (1945) found that the increased consumption of water usually following the ingestion of toxic doses of salt has a tendency to produce a progressive edema. This is reflected in the kidneys by glomerular hypertrophy together with new formation of loops and lobules. In some cases, a well-defined cardiac hypertrophy may be developed.

Ducks are apparently more susceptible to sodium chloride poisoning than chickens. Shaw (1929) reported that less than 5 grams of salt were nontoxic for 600- to 800-gram ducks, while larger doses were lethal. The time required to produce death decreased with increased dosage. Torrey and Graham (1935) reported four consecutive doses of 4 to 6 grams of salt proved fatal to half-grown Pekin ducks. Experimental birds tolerated doses of 1 to 2 grams of sodium chloride for a period of 29 days. The clinical symptoms included depression, loss of appetite, incoordination, progressive weakness, prostration, and death. The pathologic changes vary greatly. A slight congestion of the anterior part of the small intestine is the only significant lesion observed in some cases. Other individual birds may show a definite enteritis together with degenerative changes of the heart, liver, and kidneys.

Edwards (1918), who carried on extensive investigations on the toxity of sodium chloride for pigeons, reported that pigeons fed more than 3 grams per kilogram body weight showed toxic reactions. Doses in excess of 3.33 grams per kilogram body weight proved fatal in most cases. The symptoms included loss of appetite, abnormal thirst, progressive depression and weakness, followed by death. Acute congestion of the mucous membranes of the crop and proventriculus were observed. According to Buckley et al. (1939) there is no effective treatment for sodium chloride poisoning in birds.·

**Kamala.** Kamala must be regarded as a poison even though it is used as an anthelmintic for the removal of tapeworms in poultry. It is a powerful

irritant in the gastrointestinal tract. Care must be used in the administration of kamala in order to avoid toxic reactions. Following flock treatment with kamala, the egg production invariably is reduced for some time. Hall and Shillinger (1926) considered a 15-grain dose as an effective anthelmintic for mature fowls. Turkeys are less tolerant to kamala than chickens according to the report of Beach (1930). Hawn (1933) found kamala was neither a safe nor an efficient anthelmintic for turkeys. Cram (1928) warns against the use of kamala in birds affected with complicating diseases because of resulting high mortality.

**Strychnine.** Strychnine is a powerful toxic alkaloid occurring in the seeds of certain species of the Loganiaceae. It is readily absorbed from the digestive tract into the blood stream and is a powerful stimulant to the central nervous system. Its elimination from the tissues is slow, thus intensifying the cumulative action. Toxic doses produce tetanic spasms, paralysis, respiratory failure, and death. Strychnine is used extensively in the control of rodents. Accidental strychnine poisoning occasionally occurs in animals and birds consuming poisoned baits. Fowls apparently are more resistant to strychnine than mammals. According to Heinekamp (1925) the toxic dose in fowls depends largely on the quantity and nature of the crop contents; the absorption of the toxic agent being inversely proportional to the amount of food in the crop, and directly proportional to its fluidity. Gallagher (1919) regarded 0.08 gram per kilogram of body weight as the lethal dose for chickens.

**Sulfanilamide.** This drug and other closely related compounds have been used experimentally in an effort to find an effective means of controlling avian coccidiosis. During the course of these investigations it was found by Levine (1939) that sulfanilamide fed in concentrations of 0.2, 0.3, and 0.4 per cent by weight of mash for a period of two weeks was definitely toxic for chickens. He also reported that the toxicity was directly proportional to its concentration in the feed.

**Alpha naphthyl thiourea.** This preparation, commercially known as ANTU, which was recently developed as a rodenticide and one of the most effective agents for this purpose, is also highly toxic for domesticated animals and poultry. While there is much critical work to be done in determining the toxicity and lethal dosages of this product, Anderson and Richter (1946), through their experimental work with chickens, found that young chicks are quite susceptible to poisoning by this preparation. It is apparently less toxic for older birds. Chicks ranging in age from three to five weeks fed 2 per cent to 3 per cent ANTU in mash showed toxic reactions in a short time. Half of these birds were dead in 18 hours. Most of the survivors which were fed plain mash recovered and those which continued to be fed on the toxic ration ate very little and died within 4 days. The clinical symptoms include

depression, loss of appetite, listlessness, incoordination, weakness, prostration, and death. Well-defined pathologic lesions were not developed in all the poisoned birds. There was evidence of edema in the lungs and excessive quantity of fluid in the pericardial sac. Some cases showed evidence of fatty degeneration of the liver and kidneys.

Pullets averaging 1½ pounds were given doses up to the quantity to be found in 6 ounces of 2 per cent poisoned mash. Some died, and the survivors lost weight and would have been unprofitable birds if raised to maturity. Pulmonary edema, fatty degeneration of the liver and kidneys, and in some cases, degeneration of the heart, constituted the principal pathologic changes.

**Sodium monofluoracetate.** This chemical compound, commonly designated as "Compound 1080," is one of the most effective rodenticides recently developed. It is effective in the extermination of rodents but is also toxic for domesticated animals and birds. Recently conducted experiments indicate that the minimum lethal dose for chickens is about 14 milligrams per kilogram of body weight and that repeated sub-lethal dosages will produce death in a short time. The clinical symptoms include restlessness followed by a definite heart acceleration and increased respiration. Sublethal dosages frequently produce a definite blanching of the comb and wattles while heavier dosages will induce congestion and cyanosis of those appendages. As the toxemia progresses, the birds become weak and comatose. Some cases develop nervous symptoms and convulsions prior to death. The pathologic changes observed in this type of poisoning include distention of the pericardial sac with clear straw-colored fluid, hemorrhages in the cardiac tissue as well as on the endocardium, severe degeneration of the heart tissue, dark tarlike color of the blood, and large quantities of serous straw-colored fluid in the lungs and thoracic cavity. The liver appears dark in color, and the gall bladder is distended with bright green, watery bile. Degenerative changes in the kidneys may occur in some cases as well as a mild enteritis. The toxic action is primarily on the heart and no effective treatment for this condition is known at the present time.

**DDT (dichloro-diphenyl-trichlorethane).** Dry DDT crystals and the water-dispersible preparations used in reasonable quantities as indicated for their use in insect control are incapable of causing toxic reactions in domesticated poultry. From the results of experimental studies, it would appear that great quantities of 5 per cent to 10 per cent DDT preparations would have to be ingested, inhaled, or dusted upon the bird to cause an unfavorable reaction. This would be a far greater quantity than that to which poultry is likely to be exposed under normal conditions. According to Kingscote and Jarvis (1946), the oil preparations are regarded as more dangerous, but not sufficiently so as to exclude their use if used with reasonable care. Its primary use

will be for controlling gnats, flies, and some intermediate hosts of poultry parasites. McNeil and Hinshaw (1947) reported that water emulsions were more effective than kerosene emulsions and safer to use. There apparently was no advantage in using concentrations greater than 2 pounds per 100 gallons of water. The amount recommended is 5 gallons per 500 square feet, resulting in the dispersion of about 100 milligrams of powder per square foot.

The clinical symptoms observed in DDT poisoning include dyspnoea, rapid breathing through the mouth, rapid blinking of the eyelids, muscular spasms, progressive incoordination, prostration, and death. Young birds may die in a short time without showing any well-defined clinical symptoms. There are no characteristic gross pathologic changes in the internal organs except the heart which frequently shows petechial hemorrhages on the base. The majority of the birds show no specific post-mortem lesions which can be of diagnostic value.

**Carbon monoxide.** Severe losses in chicks and turkey poults may be caused by carbon monoxide poisoning, primarily due to poorly ventilated brooders or the result of defective coal or oil heating units. Adequate ventilation of brooders and housing facilities is an important factor in poultry management. Accumulation of carbon monoxide may prove fatal to entire units of young birds before the condition is discovered. The symptoms of acute carbon monoxide poisoning include restlessness, drowsiness, stupor, labored breathing, and incoordination. As the toxemia progresses the birds gasp, fall, and lie on their sides with heads thrown back. They commonly develop spasms or convulsions prior to death. Many acutely affected birds recover when removed to fresh air. In subacute cases, the feathers may appear rough, the appetite is diminished, and evidence of nutritional disturbances is manifested by retarded development and growth. Stiles (1940) found that 0.04 to 0.05 per cent carbon monoxide is sufficient to produce definite toxic reactions. The principal lesion observed in acute carbon monoxide poisoning is the bright cherry-red color of the lungs and blood. The lesions may be somewhat confusing in subacute cases and are insufficient for accurate clinical diagnosis. Such cases necessitate laboratory tests.

**Miscellaneous drugs and chemicals.** Numerous other drugs and chemicals which are known to be more or less toxic to fowls have been reported to have caused losses in both domesticated and wild birds. Some of the more common products include sodium fluoride; barium salts; chlorides of calcium and magnesium; compounds of thallium, selenium, and zinc; salicylic acid; iodine; ipecac; tartar emetic; carbolic acid; and gasoline. Poisonings by these agents are comparatively rare, and in most instances there is little accurate information regarding their toxic or lethal dosages for birds.

## PHYTOTOXINS

Phytotoxins may be considered as any toxic substance derived from plants, including the roots, stems, leaves, flowers, and seeds. The seeds and foliage are usually the plant constituents consumed by birds and are responsible for poisoning if toxic. Some plants are toxic throughout the entire growing season; others develop toxic properties only during certain stages of their development. There is also a group of plants which produces highly toxic seeds. The majority of the toxic plants are comparatively unpalatable and are naturally avoided by birds. In the absence of an abundance of succulent feed on the range, fowls frequently consume sufficient quantities of toxic foliage and seeds to cause poisoning.

**Black locust.** The black locust (*Robinia pseudoacacia*) is one of the plants whose foliage is toxic during a certain stage of its development. Barnes (1921) reported that the leaves of this plant were toxic only from about July 1st to the middle of August. He reported losses in mature fowls following the ingestion of black locust leaves. The toxic material produces a severe hemorrhagic enteritis followed by depression, paralysis, and death in from 12 to 24 hours.

**Corn cockle.** The corn cockle (*Argostemma githago*) is a weed which grows in wheat fields throughout the world. The seeds of this plant are highly toxic. The whole cockle seed is very unpalatable and is usually avoided by birds, but in ground grain mixtures it may be consumed in sufficient quantities to produce a fatal toxemia. Quigley and Waite (1931) found the toxic dose to be about 0.2 per cent of the body weight and the minimum lethal dose 0.25 per cent of the body weight of fowls. Heuser and Schumacher (1941) reported that 5 per cent of the ration or 0.3 per cent of the body weight was toxic for chickens six to ten weeks old. They found that a tolerance to the poison is frequently developed so that 0.4 to 0.5 per cent body weight could be consumed without materially affecting the growth. Ten per cent of the ration or 0.8 per cent body weight was lethal to some of their experimental fowls.

The clinical symptoms observed are a decided decrease in the respiration and heart rate, caseous lesions on the mucous membranes of the mouth, and diarrhea, the severity of which depends on the amount of toxic feed consumed. The lesions include yellow caseous exudate on the lining of the crop and varying degrees of gastroenteritis. Accumulations of clear amber fluid under the serosa of the digestive tract and in the pericardium, hemorrhages, and congested areas on the heart may be observed. Various degrees of congestion may be encountered together with degenerative changes in the liver.

**Cottonseed meal.** The active principle of cottonseed products which is toxic to both animals and birds is gossypol. Cottonseed meal has been used

as a protein supplement in stock feed for many years, and if properly used in a mixed grain ration is a valuable and economic protein supplement. Excessive quantities, however, produce toxic effects often terminating in death. Kaupp (1933) studied the toxic properties of cottonseed meal for poultry. He concluded that toxic effects were soon observed in birds consuming 1 ounce of cottonseed meal daily or its equivalent in gossypol. Clinical symptoms of generalized toxemia appear with loss of appetite, cyanosis, and emaciation followed by death in a few days after the appearance of the first clinical symptoms. The lesions include cyanotic appearance of the comb and wattles, varying degrees of gastroenteritis, and degenerative changes in the liver and kidneys.

**Coyotillo.** Losses in poultry have been reported by the consumption of the fruit and seed of the coyotillo plant (*Karwinskia humboltiana*). This plant is indigenous to southwestern Texas and Mexico. Marsh and associates (1928) reported that the clinical symptoms of this form of poisoning do not appear for several days and may appear as late as three weeks after the ingestion of the toxic substance. The toxic dose for chickens was found to be 0.3 per cent or more of the live weight, fed as dried fruit or seed. Symptoms of generalized toxemia appear followed by progressive paralysis and death.

**Crotalaria seed.** The seed of the crotalaria plant (*Crotalaria spectabilis*) contains a toxic alkaloid known as monocrotolin which, according to Thomas (1934), is highly toxic for chickens, quail, and doves. These seeds are apparently unpalatable and under ordinary conditions are not voluntarily selected as food. Turkeys apparently are more resistant to its toxic action. Poisoning may occur in either acute or chronic form, terminating fatally in from 1 day to several months. The presence of the seeds in the crop and gizzard together with the odor of crushed crotalaria leaves aids in diagnosing acute cases.

The symptoms in acute cases are distention of the crop, congested comb and wattles, and watery discharges from the nose and mouth. In chronic cases, the cyanotic color of the comb disappears, and it becomes scaly, feathers become ruffled, and diarrhea and progressive weakness are manifested prior to death. The characteristic lesions described by Emmel (1937) include numerous petechiae in the serous membranes and visceral fat (Fig. 39.4). The liver has a dark brown appearance and is usually mottled. Urates accumulate in the kidneys and ureters.

*Crotalaria retusa* was found to be about as toxic for birds as *C. spectabilis*. Other species of crotalaria, including *C. striata*, *C. grantiana*, *C. intermedia*, and *C. incana*, were found to be nontoxic for chickens and quail when they were fed considerable quantities of the seeds under experimental conditions.

**Daubentonia seeds.** The Daubentonia (*Daubentonia longifolia*), also

called the Sesbania, is a native of Mexico but was introduced to the southern states as an ornamental shrub. The seeds of this plant are readily eaten by poultry and are extremely toxic. According to Shealy and Thomas (1928), the ingestion of as few as nine seeds will cause death in birds. The first clinical symptoms observed are a staggering gait, accompanied by drooping of the wings. Depression, general debility, and unthriftiness soon become apparent. The comb becomes cyanotic and the head may hang over to one side. Muscular twitching, diarrhea, emaciation, and extreme weakness is followed by death in 24 to 72 hours after the appearance of the first symptoms. The lesions include severe gastroenteritis with ulceration of the proventriculus and gizzard together with degenerative changes of the liver.

**Death camas.** The death camas belong to the genus Zygadenus, and the members of this genus, according to Marsh *et al.* (1915), generally conceded to be poisonous are *Z. glaberrimus, Z. intermedius, Z. mexicanus, Z. nuttallii, Z. paniculatus,* and *Z. venenosus.* Niemann (1928) reported an outbreak of poisoning in the domestic fowl due to the ingestion of *Z. nuttallii.* This plant is not very palatable and is only eaten by fowls on the range in the early spring and late fall when other green feed is comparatively scarce. Experimental



Fig. 39.4. Hemorrhages of the epicardium and myocardium in acute *Crotalaria spectabilis* seed poisoning in a chicken. (Emmel, Jour. A.V.M.A.)

feedings of 5 to 10 grams to chickens produced marked clinical symptoms in 12 hours including salivation, incoordination, muscular weakness, and diarrhea followed by prostration and death. No definite lesions are associated with this type of poisoning. A strong, penetrating, disagreeable odor from the internal organs was noted, and muscular atrophy was observed. The mesenteric and abdominal blood vessels appeared congested. The lumen of the digestive tract was noticeably diminished in size.

**Glottidium seed.** *Glottidium vesicarium* (Jacq.) Harper is quite common along the coastal plain from North Carolina to Florida and Texas, having been introduced from the West Indies. The seeds of this plant are toxic for poultry. Under ordinary conditions fowls do not select these seeds as food but if underfed may consume sufficient quantities to be toxic. Emmel (1935)

produced toxic effects by experimental feeding of *G. vesicarium* seeds to fowls. The clinical symptoms in acute poisoning are prostration, and cyanotic appearance of comb and wattles accompanied by diarrhea. In chronic cases the feathers become ruffled, copious yellow diarrhea persists, and the birds may become emaciated. The combs appear light in color and become scaly. The most characteristic lesions observed include necrotic enteritis, necrotic areas in the lining of the gizzard, and degenerative changes in the liver and kidneys.

**Vetch seed.** Reports of toxicity as the result of feeding large quantities of vetch seed to chickens are recorded in the literature. Little reliable information can be secured on the toxic principles of the vetches from the reports published either in this country or abroad. Numerous incidents of poisoning, commonly referred to as "lathyrism," are recorded, in which birds show severe central nervous disturbances including spasmodic convulsions, paresis, and death following the use of large quantities of Lathyrus peas in the feed. Stockman (1931) regarded the nature of this toxic reaction highly controversial. Anderson and his associates (1925) indicated that the seed of *Lathyrus sativus* is not toxic unless contaminated by seeds of a variety of vetch, *Vicia sativa* L., var. *Augustifolia*. Horvath (1945) reported that the vetchling (*Lathyrus cicera*), when fed as a sole food, seemed to exert a toxic effect on hens, resulting in a loss of weight. No characteristic pathologic lesions were produced. Several varieties of vetch seed contain a cyanogenic glucoside "vicianin" which is decomposed by an enzyme (vicinase) into hydrocyanic acid, benzaldehyde, and a disaccharide (vicianose). It is possible that large quantities of such feed could cause considerable loss in a flock of birds.

**Milkweed.** Two of our common species of milkweed, *Asclepias tuberosa* and *A. incarnata,* contain the bitter glucoside asclepidin which apparently is toxic to animals and birds. Pammel (1911) includes *A. vestita* and *A. mexicana* in the list of toxic species of the milkweed family. Campbell (1931) reported serious losses in poultry caused by the consumption of the narrow-leaved, whorled milkweed, *A. mexicana*. Experimental investigations indicated that all parts of the plant are toxic. Pammel (1917) reported milkweed poisoning in chickens resulting in the loss of approximately 500 birds. Stiles (1942) reported extensive losses in turkey poults resulting from the consumption of whorled milkweed (*Asclepias galioides*). Poultry, as well as other species of livestock, are not likely to eat milkweed except when it is the only succulent feed available.

The clinical symptoms may vary considerably, depending on the quantity of the toxic material eaten. The first symptom observed was lameness which developed rapidly into complete loss of muscular control. The neck became

twisted and the head drawn back. The affected birds often lay on their sternums or sat on their hocks alternately extending and retracting their heads at frequent intervals. This condition did not seem to be a true paralysis but appeared to be an overstimulation of the motor nerve centers with complete loss of coordination. At times the birds would fall over and struggle with violent convulsive movements of the legs. In some instances the symptoms gradually subsided, followed by recovery. In fatal cases the symptoms became progressively worse, followed by prostration, coma, and death. No characteristic lesions were found upon autopsy.

**Nightshade.** The black nightshade (*Solanum nigrum*) is a common weed in yards and poor pasture land. The immature fruit of this plant contains the alkaloids solanin and solanidin which are toxic to man and animals. The toxicity of the plants is believed to be influenced by the soil, climate, and degree of plant maturity. Hansen (1925) reported fatal poisoning in chickens and ducks attributed to black nightshade. The clinical symptoms include incoordination, prostration, paralysis, and death. The pupils of the eyes may be dilated. No characteristic lesions are reported except evidence of severe toxemia.

**Lily of the valley and oleander.** The flowers, leaves, and stems of the lily of the valley (*Convallaria majalis*) and the leaves of the oleander (*Nerium oleander*) were reported by Bardosi (1939) to be poisonous for geese, ducks, and hens. The lily of the valley flowers were found to be lethal to geese in doses of 13 grams and to ducks in doses of 12 grams. Doses of 30 grams produced only a mild enteritis in mature chickens.

The dried leaves of the oleander plant of the previous season's growth proved fatal to geese in 24 hours after the ingestion of 6 grams. The lethal dose for ducks was found to be 3 grams. The young leaves of this plant proved fatal to hens in doses of 15 grams. Hinshaw reported losses in turkey poults within 24 hours as the result of eating young shoots of oleander. The post-mortem examination of the poults showed hemorrhagic enteritis. (See chapter on Diseases of the Turkey.)

The clinical symptoms of oleander poisoning include general depression, weakness, diarrhea, accelerated heart rate, impaired vision, muscular incoordination, and in some instances paralysis of the wings. Various degrees of gastroenteritis and liver degeneration occur in fatal cases.

**Potatoes.** Under certain conditions the potato (*Solanum tuberosum*) is poisonous to domesticated animals and poultry. Greened tubers produced by exposure to light and young potato sprouts contain considerable amounts of the alkaloid solanin which is highly toxic. Analyses have shown that the solanin content of the sprouts and peelings is higher than that of the interior of the tuber. Hansen (1927) reported several outbreaks of poisoning in

poultry resulting from the ingestion of potato sprouts. Losses occurred within a few hours after the consumption of the toxic sprouts. Temperton (1944) reported losses in ducks as the result of eating either cooked or uncooked sprouted potatoes. This type of poisoning is similar to that of poisoning by other plants of the nightshade family. However, where large amounts of potatoes are fed to poultry, it is advisable as a safety measure to cook green or sprouted potatoes and to discard the residual water. From the standpoint of nutrition, raw potato starch is poorly digested by poultry, and cooking will result in a more efficient utilization of this type of feed.

**Tobacco.** The tobacco plant (*Nicotiana tobacum* L.) contains the toxic alkaloid nicotine. Hunter and associates (1931, 1934) reported that growing chicks over three weeks of age can tolerate as much as 0.06 per cent nicotine in the ration without any toxic reaction. The feeding of the same nicotine levels in the form of ground cigar clippings having only 0.86 per cent nicotine content retarded the growth and development of chicks, causing some losses. Toxic doses of tobacco produce similar reactions to those of nicotine sulfate poisoning.

**Algae.** Certain types of algae, including the *Microcystis aeruginosa*, which grow abundantly in lakes under certain conditions, may become concentrated in localized areas by the action of strong winds blowing the surface of the water in one direction for a number of days. Great quantities of algae are deposited on the banks and in the shallow waters along the shore line. The disintegration of this material produces toxins which are reponsible for the loss of various species of wild life as well as domesticated animals and birds. This condition has frequently been called "Water Bloom." Fitch and his associates (1929) reported losses of livestock from this cause in Minnesota. Brandenburg and Shigley (1947) reported this condition in North Dakota. It has also been reported in northern Iowa and in parts of Canada. It usually occurs in the latter part of July, August, and September. The exact nature of the toxin is not known at the present time. Apparently, the toxin is an intermediate product of disintegration, as these toxic properties disappear during the latter stages of decomposition.

The toxicity of this material is directly proportional to the concentration. Some waters taken from the shores of lakes are extremely toxic to birds and animals. Under experimental conditions, oral dosages of 10 cc. to 30 cc. will produce death in mature ducks and chickens in 10 to 45 minutes. The toxin is thermostable and is affected little, if any, by boiling. The symptoms include restlessness, twitching of muscles, nervous manifestations, spasms, convulsions, paralysis, and death. These clinical manifestations resemble strychnine poisoning in many respects. The post-mortem lesions include generalized cyanosis; dark, tar-colored blood; liver congested and dark in color; heart dilated and distended; muscles also congested and dark in color. No

characteristic hemorrhages are observed. Ashworth and Mason (1946) described in detail the symptoms and lesions associated with algae poisoning in laboratory animals.

Because of the highly toxic nature of this material and the rapidity with which it acts, there are no therapeutic measures effective. Poultry should be restricted to areas free from toxic material.

Nontoxic algae may be responsible for considerable losses in young chickens. The mature birds are seldom affected, but the young chicks which wade out along the shore line and feed on insects alighting on the floating scum or masses of algae invariably swallow various amounts of this material. This scum becomes lodged in the nostrils as well as the digestive and respiratory passages, causing strangulation and suffocation. Symptoms characteristic of strangulation are observed. Post-mortem examination reveals obstruction of the nostrils and respiratory passages as well as those of the digestive tract.

Losses of poultry associated with the ingestion of algae should be investigated at once. The toxicity of algae can be determined readily by injecting 2 cc. or more of a sample filtered through gauze, intraperitoneally into laboratory animals or chickens. Toxic material will produce typical clinical symptoms in a short time and death within an hour or so. A prompt determination of toxicity may prevent serious losses to all species of livestock.

## INSECTS

Insects of various kinds annoy poultry and act as intermediate hosts for poultry parasites, but only few of those eaten by birds are considered poisonous. Only one insect is described which, if eaten in sufficient numbers, produces a severe toxic reaction in chickens, i.e., the rose chafer.

**Rose chafer.** Rose chafers (*Macrodactylus subspinosus*) are abundant during the latter part of May and June, and early July in Canada and the eastern United States extending as far west as Colorado. The toxic properties of this insect for chickens have been reported by Bates (1916), Gallagher (1920), and Lamson (1916, 1922). Fatal cases of poisoning in young chickens of various ages have been recorded, but mature fowls are seldom killed. Chickens will feed ravenously upon these insects if available, and fifteen to twenty rose chafers are sufficient to kill a chicken one week old while birds about three weeks of age show a toxic reaction after eating twenty-five to forty-five of these insects. A fatal reaction usually results in 24 hours, or the birds gradually recover. Watery extracts made from crushed rose chafers proved toxic when administered to chickens. Lamson was of the opinion that the poisonous principle is a neurotoxin which has a direct effect on the heart action.

The clinical symptoms include drowsiness, incoordination, weakness,

prostration, convulsions, and retraction of head and neck over the back of the affected chicken. Death usually occurs in less than 24 hours subsequent to eating rose chafers or from $\frac{1}{2}$ to 1 hour after the appearance of the first symptoms. The post-mortem examinations fail to show characteristic lesions other than injection of the blood vessels of the heart in some cases.

## MISCELLANEOUS FOOD POISONS

**Selenium.** Toxic reactions from eating grains grown in certain limited areas due to specific toxic mineral constituents have been reported by Franke and associates (1934) in parts of South Dakota. The so-called alkali disease was found to be due to the high selenium content of the grain grown in that locality. Losses of livestock were reported by Moxon (1937) from selenium poisoning. Studies at the South Dakota Experiment Station indicated that toxic grains fed at levels which contained 15 parts per million of selenium to laying hens resulted in reduced weight, caused a decided reduction in egg size, and practically destroyed the hatchability. According to Poley and associates (1937) the feeding of 5 parts per million of selenium did not appreciably affect the hatchability even though some evidence of selenium poisoning was apparent.

**Protein poisoning.** Protein poisoning in poultry may be both quantitative and qualitative. Jull (1930) points out the harmful effects of excessive protein in the ration. The clinical symptoms caused by excessive amounts of protein are those of generalized toxemia including depression, leg weakness, prostration, and coma followed by death. This problem is primarily one of management, and losses from this cause can be prevented by using properly balanced rations.

Birds may also be poisoned by the ingestion of proteins of poor quality such as partly decomposed foods. Decomposed proteins frequently prove toxic if fed in sufficient amounts. It is believed that the products of disintegration are responsible for the toxic reaction, which is not necessarily associated with bacterial toxins produced as in the case of botulism.

Feeds of various kinds, whether they are proprietary feed mixtures or some ingredient of home-mixed rations, are often suspected of being responsible for poultry losses. Quigley and Waite (1931) reported that investigations on numerous samples examined over a period of three years failed to show any toxic reaction by actual feeding trials. Various other reports indicated that comparatively few feeds suspected of being poisonous proved to be so when examined.

The losses in poultry as the result of poisoning from any cause are exceedingly small compared to the losses attributed to infectious diseases, parasites, and other causes. Positive diagnoses of poisoning depend entirely upon the

discovery of the poison in the bird by chemical analyses or by the detection of a specific poison in the food supply.

## REFERENCES

Anderson, L. A. P., Howard, A., and Simonsen, J. L.: 1925. Studies on lathyrism. Indian Jour. Med. Res. 12:613.

Anderson, W. A., and Richter, C. P.: 1946. Toxicity of alpha naphthyl thiourea. Vet. Med. 41:302.

Ashworth, C. T., and Mason, M. F.: 1946. Observations on the pathological changes produced by a toxic substance present in blue-green algae (*Microcystis aeruginosa*). Am. Jour. Path. 22:369.

Barber, P. G., and Hubster, E. B.: 1933. Arsenic poisoning in poultry. Vet. Med. 28:500.

Bardosi, Z.: 1939. Toxicity of lily of the valley and oleander leaves for fowls (trans. title). Thesis, Budapest, Abst. Vet. Bul. (1940) 10:624.

Barnes, M. F.: 1921. Black locust poisoning of chickens. Jour. Am. Vet. Med. Assn. 59:370.

Bates, J. M.: 1916. The poisonous character of rose chafers. Science 43:209.

Beach, J. R.: 1930. Intestinal worms of poultry. No. Am. Vet. 11:45.

Beaudette, F. R., Hudson, C. B., and Weber, A. L.: 1933. Phosphorous poisoning in poultry. No. Am. Vet. 14:39.

Bleecker, W. L., and Smith, R. M.: 1933a. Further studies on the relative efficiency of vermifuges for poultry. Jour. Am. Vet. Med. Assn. 83:76.

———— and Smith, R. M.: 1933b. Nicotine sulfate as a vermifuge for the removal of ascarids from poultry. Jour. Am. Vet. Med. Assn. 83:645.

Bradenburg, T. O., and Shigley, F. M.: 1947. "Water Bloom" as a cause of poisoning in livestock in North Dakota. Jour. Am. Vet. Med. Assn. 110:384.

Buckley, J. S., Bunyea, H., and Cram, E. B.: 1939. U.S.D.A., Farmer's Bul. 1652.

Campbell, H. W.: 1931. Poisoning in chickens with whorled milkweed. Jour. Am. Vet. Med. Assn. 79:102.

Carpenter, C. D.: 1931. The use of nicotine and its compounds for the control of poultry parasites. Jour. Am. Vet. Med. Assn. 78:651.

Cooley, R. A., Parker, J. R., and Strand, A. L.: 1923. Improved methods of controlling grasshoppers. Mont. Agr. Exper. Sta., Circ. 112.

Cram, E. B.: 1928. The present status of our knowledge of poultry parasitism. No. Am. Vet. 9:43.

Edwards, J. T.: 1918. Salt poisoning in pigs and poultry. Jour. Comp. Path. and Therap. 31:40.

Emmel, M. W: 1935. The toxicity of *Glottidium vesicarium* (Jacq.) Harper seeds for the fowl. Jour. Am. Vet. Med. Assn. 87:13.

————: 1937. The pathology of *Crotalaria spectabilis* Roth seed poisoning in the domestic fowl. Jour. Am. Vet. Med. Assn. 90:627.

Fitch, C. P., Bishop, L., and Boyd, W. L.: 1929. "Water Bloom" as a cause of poisoning in domestic animals. Cornell Vet. 21:30.

Franke, K. W., Rice, T. D., Johnson, A. G., and Schoening, H. W.: 1934. Report on a preliminary field survey of the so-called "alkali disease" of livestock. U.S.D.A., Circ. 320.

Gallagher, B. A.: 1919. Experiments in avian toxicology. Jour. Am. Vet. Med. Assn. 54:337.

————: 1920. Rose-chafer poisoning in chickens. Jour. Am. Vet. Med. Assn. 57:692.

————: 1924. Canned goods preserved with boric acid poisonous to chickens. No. Am. Vet. 5:125.

Guberlet, J. E.: 1922. Potassium nitrate poisoning in chickens with a note on its toxicity. Jour. Am. Vet. Med. Assn. 62:362.

Hall, M. C., and Shillinger, J. E.: 1926. Kamala, a satisfactory anthelmintic for tapeworms in poultry. No. Am. Vet. 7:51.

Hansen, A. A.: 1925. Nightshade poisoning in chickens and ducks. Jour. Am. Vet. Med. Assn. 66:502.

————: 1927. Stock poisoning by plants in the nightshade family. Jour. Am. Vet. Med. Assn. 71:221.

Hanzlik, P. J., and Presho, E.: 1923. Comparative toxicity of inorganic lead compounds and metallic lead for pigeons. Jour. Pharmacol. and Exper. Therap. 21:123.

Hare, T., and Orr, A. B.: 1945. Poultry poisoned by zinc phosphide. Vet. Record 57:17.

Hawn, M. C.: 1933. The value of kamala as a tenicide for young turkeys. Jour. Am. Vet. Med. Assn. 83:400.

Heinekamp, W. J. R.: 1925. The resistance of fowl to strychnine. Jour. Lab. and Clin. Med. 11:209.

Heuser, G. F., and Schumacher, A. E.: 1941. The feeding of corn cockle to chickens. Poultry Sci. 20:463.

Horvath, A. A.: 1945. Toxicity of vetch seed for chickens. Poultry Sci. 24:291.

Hudson, C. B.: 1936. Naphthalene poisoning in poultry. Jour. Am. Vet. Med. Assn. 89:219.

Hunter, J. E., and Haley, D. E.: 1931. The effect of various concentrations of nicotine in tobacco on the growth and development of fowls. I. A study of the nicotine tolerance of growing chicks. Poultry Sci. 10:61.

————, Haley, D. E., and Knandel, H. C.: 1934. Effect of concentrations of nicotine on growth and development. II. Growth and development of chicks as influenced by the addition of ground tobacco to the ration. Poultry Sci. 13:91.

Johns, F. M.: 1934. A study of punctate stippling as found in the lead poisoning of wild ducks. Jour. Lab. and Clin. Med. 19:514.

Jull, M. A.: 1930. Poultry Husbandry. McGraw-Hill Book Co., Inc., New York. P. 328.

Kaupp, B. F.: 1933. Poultry Diseases. 6th Ed. Alexander Eger, Chicago, Ill. P. 444.

Kingscote, A. A., and Jarvis, C. H.: 1946. Report on experiments conducted to establish the tolerance of turkeys to DDT. Canad. Jour. Comp. Med. 10:211.

Krakower, C. A., and Goettsch, M.: 1945. Effect of excessive ingestion of sodium chloride on the chick, with particular reference to renal changes. Arch. Path. 40:209.

Lamson, Jr., G. H.: 1916. The poisonous effects of the rose chafer upon chickens. Science 43:138.

————: 1922. The rose chafer as a cause of death of chickens. Storrs Agr. Exper. Sta., Bul. 110:115.

Lander, C. D.: 1926a. Veterinary Toxicology. Alexander Eger, Chicago, Ill. P. 57.

————: 1926b. Ibid., p. 75.

Levine, P. P.: 1939. The effect of sulfanilamide on the course of experimental avian coccidiosis. Cornell Vet. 29:309.

Marsh, C. D., Clawson, A. B., and Marsh, H.: 1915. Zygadenus or death camas. U.S.D.A., Bul. 125.

————, Clawson, A. B., and Roe, G. C.: 1928. Coyotillo (Karwinskia humboldtiana) as a poisonous plant, U.S.D.A., Tech. Bul. 29.

McNeil, E., and Hinshaw, W. R.: 1945. Effects of mercuric chloride on turkeys and on Hexamita meleagridis. Poultry Sci. 24:516.

———— and Hinshaw, W. R.: 1947. Experience with DDT on a turkey ranch. Vet. Med. 42:181.

Moxon, A. L.: 1937. Alkali disease or selenium poisoning. S. D. Agr. Exper. Sta., Bul. 311.

Niemann, K. W.: 1928. Report of an outbreak of poisoning in the domesticated fowl, due to death camas. Jour. Am. Vet. Med. Assn. 73:627.

Nunn, J. A.: 1907. Veterinary Toxicology. Baillière, Tindall, and Cox, London. P. 73.

Pammel, L. H.: 1911. A Manual of Poisonous Plants. The Torch Press, Cedar Rapids, Ia. P. 696.

————: 1917. Milkweed poisonous to chickens. Am. Jour. Vet. Med. 12:236.

Parker, S. L.: 1929. Effects of early handicaps on chickens as measured by yolk absorption and body weight to twenty weeks of age. Hilgardia 4:1.

Poley, W. E., Moxon, A. L., and Franke, K. W.: 1937. Further studies of the effects of selenium poisoning on hatchability. Poultry Sci. 16:219.

Pullar, E. M.: 1940a. The toxicity of various copper compounds and mixtures for domesticated birds. Australian Vet. Jour. 16:147.

————: 1940b. The toxicity of various copper compounds and mixtures for domesticated birds. 2. Australian Vet. Jour. 16:203.

Quigley, G. D., and Waite, R. H.: 1931. Miscellaneous feeding trials with poultry. Md. Agr. Exper. Sta., Bul. 325:343.

Shaw, P. A.: 1929. Duck disease studies. II. Feeding of single and mixed salts. Proc. Soc. Exper. Biol. and Med. 27:120.

Shealy, A. L., and Thomas, E. F.: 1928. Daubentonia seed poisoning of poultry. Univ. Fla. Agr. Exper. Sta., Bul. 196.

Sherwin, C. P., and Crowdle, J. H.: 1922. Detoxication in the organism of the fowl. Proc. Soc. Exper. Biol. and Med. 19:318.

Stiles, G. W.: 1940. Carbon monoxide poisoning of chicks and poults in poorly ventilated brooders. Poultry Sci. 19:111.

———: 1942. Poisoning of turkey poults from whorled milkweed (*Asclepias galioides*). Poultry Sci. 21:263.

Stockman, R.: 1931. The poisonous principle of Lathyrus and some other leguminous seeds. Jour. Hyg. 31:550

———: 1934. The chemistry and pharmacology of Lathyrus peas. Jour. Hyg. 34:145.

Temperton, H.: 1944. Effect of green and sprouted potatoes on laying pullets. Vet. Med. 39:13.

Thomas, E. F.: 1934. The toxicity of certain species of crotalaria seed for the chicken, quail, turkey, and dove. Jour. Am. Vet. Med. Assn. 85:617.

Torrey, J. P., and Graham, R.: 1935. A note on experimental salt poisoning in ducks. Cornell Vet. 25:50.

Van Zyl, J. P.: 1929. Annual Report of the Director of Veterinary Services, Union of South Africa 15:1189.

Wetmore, A.: 1922. Lead poisoning in waterfowl. U.S.D.A., Bul. 793.

Whitehead, F. E.: 1934. The effect of arsenic, as used in poisoning grasshoppers, upon birds. Okla. Agr. Exper. Sta., Bul. 218.

Wickware, A. B.: 1945. Grasshoppers: A potential danger to turkeys. Canad. Jour. Comp. Med. 9:80.

Wilson, H. F., and Holmes, C. E.: 1936. Effect on chickens of arsenic in grasshopper bait. Jour. Econ. Ent. 29:1008.

Winchell, C. W.: 1925. The use of calcium cyanide in the killing of condemned chickens. Rural New Yorker 84:76.

Witter, J. F.: 1936. A preliminary report on the injurious effect of sodium bicarbonate in chicks. Poultry Sci. 15:256.

# DISEASES OF THE TURKEY

By W. R. HINSHAW, *Department of Veterinary Science,*
*University of California, Davis, California*

☆ ☆ ☆

## DIETARY DISEASES

The turkey responds differently to many of the nutritional factors than does the chicken, and for this reason the requirements are often different. Likewise, the pathological changes seen when these factors are lacking may vary from those seen in the chicken under similar conditions. Only the pertinent facts and differences are given in this section. For more complete information on the nutritional requirements of birds, the reader is referred to the section on nutrition, and to recent references in the literature, as for example, Ewing (1943) and Marsden and Martin (1944).

Six vitamins (A, $B_1$, D, E, G, and K) are definitely known to be needed by turkeys, according to Asmundson and Jukes (1939). Vitamins are especially important in the starting ration because poults have a higher requirement for some of the vitamins than do baby chicks.

All vitamins required by turkeys are present in the commonly used feedstuffs, and it is not necessary to purchase them in purified form or in expensive proprietary mixtures. Vitamins A, D, and G (riboflavin) are most likely to be deficient in turkey rations. Vitamins $B_1$ (thiamin) and K are both needed by turkeys, but are present in all ordinary rations to an extent which makes deficiency nearly impossible. A deficiency of pantothenic acid (filtrate factor) is the cause of dietary dermatitis in chickens but does not cause it in turkeys. Lack of this factor will, however, slow up growth. It is present in most turkey feeds in amounts that are adequate for growth. Table 1, compiled by Kratzer (1947), summarizes the symptoms of vitamin deficiencies, the amounts required for normal growth, and the principal sources of the vitamins required by turkeys. A paper by Jukes, Stokstad, and Belt (1947) gives additional data.

## VITAMIN A DEFICIENCY

Vitamin A, a fat-soluble vitamin, is found in many fish oils, in all green leaves, yellow corn, yellow carrots, and similar yellow and red root crops. Vitamin A exists in both plant and animal forms. The plant form is "carotene," an orange-red pigment. The animal form of vitamin A is nearly colorless and is present in certain fish oils. Birds have the ability to convert the plant form of vitamin A to the animal form; it does not seem to matter which form is fed to birds because they can make equally good use of either.

TABLE 1

VITAMIN REQUIREMENTS OF TURKEYS (KRATZER, 1947)

| Vitamin | Symptoms of Deficiency | Amount Required per Pound of Feed | Best Sources |
|---|---|---|---|
| A | Poor growth, unsteady gait, watery eyes, nasal discharge white cheesy exudate from eyes and sinuses, pustules in mouth and esophagus, white, flaky exudate in bursa of Fabricius, high mortality. | Poults 2500 I.U. Breeders 4000 I.U. | Fresh greens, dehydrated alfalfa, fish oils. |
| D | Poults—rickets, characterized by poor growth, leg weakness, enlarged hocks, crooked keels, beaded ribs, soft bones and beak, high mortality.<br><br>Breeders—low egg production, low hatchability, soft-shelled eggs. | Poults 450 AOAC units. Breeders 450 AOAC units. | Biologically tested fish oils, activated animal sterol. (Activated animal sterol is more active than vitamin D from fish oils when phosphorus of the ration is in organic combination. Vitamin D from codliver oil should be supplied at 900 AOAC units per pound when inorganic phosphorus of ration is low.) Direct sunlight can replace dietary vitamin D. |
| E | Not studied in turkeys (may be produced experimentally in chickens in which it results in paralysis and uncoordinated movements in chicks and poor hatchability in breeding hens). | Practical rations contain adequate amounts. | Fresh greens, grain, dehydrated alfalfa. |
| K | Blood fails to clot normally. Small injuries cause excessive bleeding. | Practical rations contain adequate amounts. | Fresh greens, dehydrated alfalfa. |
| Thiamin ($B_1$) | Not studied extensively in turkeys. (In chicks it results in poor growth, nervous symptoms including head retraction and convulsions, high mortality.) | Practical rations contain adequate amounts. | Grains, grain by-products, oil cake meals. |
| Riboflavin ($B_2$, G) | Poor growth, leg weakness, dermatitis in poults, low hatchability in breeders. | Poults 2000 micrograms. Breeders 1800 micrograms. | Milk products, liver meal, fresh greens, dehydrated alfalfa, yeast, fermentation by-products, synthetic riboflavin. |
| Pantothenic acid | Poor growth, high mortality. | Poults 6250 micrograms. | Yeast, molasses, liver meal, milk products, dehydrated alfalfa, wheat bran. |
| Pyridoxine ($B_6$) | Poor growth, uncoordinated movements, convulsions, high mortality. | Poults 2000 micrograms. | Grain, grain by-products, soybean oil meal, milk products, yeast, dehydrated alfalfa, animal products. |
| Niacin | Poor growth, inflammation of the mouth, poor feathering, perosis. | Exact requirement unknown. | Wheat and wheat by-products, yeast, liver meal, fish meal, meat scrap. |
| Biotin | Poor growth, dermatitis, perosis, high mortality. | Exact requirement unknown. | Fresh greens, dehydrated alfalfa, soybean oil meal, cane molasses, grain, grain by-products. |

TABLE 1 (continued)
VITAMIN REQUIREMENTS OF TURKEYS (KRATZER, 1947)

| Vitamin | Symptoms of Deficiency | Amount Required per Pound of Feed | Best Sources |
|---|---|---|---|
| Pteroylglutamic acid (B<sub>c</sub>, Folic acid) | Poor growth, paralysis. | Exact requirement unknown. | Green feed, dehydrated alfalfa, yeast, liver meal, wheat bran, soybean oil meal. |
| Choline | Poor growth, perosis. | Poults 900 milligrams. | Yeast, oil cake meals, fish meal, liver meal, synthetic choline. |
| Anti-gizzard erosion factor | Eroded areas in gizzard lining. | Unknown. (Factor is chemically unidentified.) | Liquid milk products, fresh greens, wheat bran. |
| Ascorbic acid (C) | Dietary source not required by turkeys. | | |
| Unknown factors | There is experimental evidence for the existence of other factors required for normal growth and reproduction in turkeys. Deficiencies are not likely to be encountered in practical rations containing a variety of natural feedstuffs. | | |

Since turkeys require considerably more vitamin A than do chickens, they are more susceptible to avitaminosis A. Hinshaw and Lloyd (1934), Scott (1937), and Wilgus (1940) have shown that turkeys require from two to four times as much vitamin A in their rations as do chickens.

**Symptoms.** Poults fed a vitamin A free diet from the time of hatching begin to show symptoms within three to four weeks, depending upon the amount stored in their bodies at the start. They appear listless, walk unsteadily, and have a tendency to sit with sagging wings, drooping heads, and closed eyes. Later symptoms include watery eyes, swelling of the third eyelid (nictitating membrane), and nasal discharge. A milky exudate, followed by a white cheesy exudate, appears in the eyes and head sinuses if the poult lives for any period after showing the first symptoms. The nictitating membrane has a dry, rough appearance, and the surface may be covered with finely divided powdery exudate. When observed early in the morning, many poults will be found with their eyes closed by a sticky exudate adhering to the lids. Figure 40.1 shows, for comparison, a poult and a chick, both exhibiting typical symptoms. The course of the disease in poults is very short, and 100 per cent mortality occurs within two weeks after symptoms appear if vitamin A is not supplied.

In older birds receiving inadequate amounts of vitamin A, the symptoms are similar but more pronounced, probably because of the chronicity of the disease. In many instances symptoms that cannot be differentiated from colds and infectious sinusitis are observed. It is, therefore, highly desirable in all outbreaks of these diseases in turkeys to eliminate the possibility of vitamin A deficiency when making a diagnosis.

**Autopsy findings.** Lesions are confined principally to the upper digestive tract and to the head. They consist of swollen caseated glands (pustules) in the posterior part of the mouth (Figs. 40.2 and 40.3), the upper esophagus, and the crop, and an involvement of the sinuses of the head (Figs. 40.4 and 40.5). The bursa of Fabricius, an accessory pouchlike organ present only in young poults, located dorsal to the rectum and having an opening into the cloaca, is usually filled with a white, flaky exudate. Urate deposits on the



Fig. 40.1. A five-week-old poult and a six-week-old chick, both showing typical symptoms of vitamin A deficiency. (Hinshaw, Univ. of Calif.)

intestines, heart, and lungs, and swollen kidneys filled with urates, common in chickens suffering from vitamin A deficiency, have not been observed in turkeys.

By means of chemical tests available in many diagnostic laboratories, it is possible to determine if turkeys have adequate storage of vitamin A in their bodies. These tests are based on the determination of pro-vitamin A or vitamin A in the livers and may prove an aid in differentiating avitaminosis A from diseases showing similar symptoms.

**Control and prevention.** Control and prevention consist of furnishing sufficient vitamin A in the ration. This can be supplied by large amounts of bulky feeds such as fresh greens and alfalfa meal. (See Tables 1 and 2.) It should be noted that the vitamin A of fish oils tends to diminish after the oil

TABLE 2.

| Feedstuff | *Approximate Units of Vitamin A per Pound | Percentage in Ration to Supply 4,500 Units per Pound |
|---|---|---|
| Yellow corn......... | 3,000 | 50 (would supply only 1,500 units) |
| Alfalfa meal, containing 45 milligrams of carotene per pound | 75,000 | 6 |
| Fresh green leaves.......... | 45,000 | 10 |
| High-potency fish oil, 3,000 A | 1,360,000 | 0.33 |

*The vitamin A unit referred to is the United States Pharmacopeia (XI) unit, which is the same as the International unit, defined as the activity of 0.6 micrograms of beta carotene of the International Standard Reference. Table taken from Asmundson and Jukes (1939).

FIG. 40.2. Portion of esophagus of a turkey hen showing pustular lesions in vitamin A deficiency. (Hinshaw, Univ. of Calif.)



FIG. 40.3. A—floor of mouth and pharyngeal region of a 40-day-old turkey that died from vitamin A deficiency. B—same of a 45-day-old chick; note the large number of pustules in B as compared with A. The specimens are typical for the two species. C—sagittal section of bursas of Fabricius and caseous plugs characteristic of vitamin A deficiency in young turkeys and chickens; the left bursa was from a poult; the right, from a chick; the middle specimens are typical caseous plugs from bursas taken from chicks. (Hinshaw, Univ. of Calif.)

has been mixed in the mash, and that, when alfalfa meal is stored, its vitamin A content decreases, especially in warm weather.

## RICKETS (VITAMIN D DEFICIENCY)

Rickets, a bone disease affecting all animals and birds, is caused by failure to receive a proper balance of vitamin D and minerals. Vitamin D is also necessary for egg production and hatchability. A lack of it contributes to the development of crooked breast bones in turkeys. This vitamin is present in certain fish oils. It is also supplied by direct sunlight, which changes certain substances in the skin to vitamin D. Vitamin D is of great importance to poults, which have a high requirement for this factor.

**Symptoms.** Leg weakness, awkwardness of gait, softness of the beak and leg bones, and ruffled, unkempt feathers are characteristic of this condition. The affected poults fail to gain weight and finally die if the balance of minerals and vitamin D is not corrected.

FIG. 40.4. Turkey hen showing typical symptoms of a vitamin A deficiency. Taken 87 days after being on a deficient ration. The hen died 3 days after the picture was taken. (Hinshaw, Univ. of Calif.)

According to Scott, Hughes, and Loy (1932), poults receiving a diet deficient only in vitamin D will develop symptoms in 18 to 20 days, and 100 per cent mortality will occur within 30 days after hatching.

**Autopsy findings.** Softness of the bony structures and beading of the ribs are the most common autopsy findings. A definite diagnosis depends on a chemical analysis of the bones or blood, or upon the "line" test for rickets.

**Prevention and control.** There are several forms of vitamin D, some of which are very effective for the prevention of rickets in rats, but not in chicks and poults. For this reason, a source of vitamin D such as fish oil should be tested with chicks before use in turkey feeding. Such a test is commonly made by the A.O.A.C.[1] procedure for determining chick units. Poults need several times as much vitamin D in their feed as chicks (see Table 1). This explains why poults sometimes develop rickets when placed on chick starting mashes.

[1] The vitamin-D chick unit referred to in this section is that determined according to the tentative procedure of the Association of Official Agricultural Chemists in which it is defined that 1 U.S.P. unit of vitamin D (in the United States Pharmacopeia Reference Cod-Liver Oil) equals 1 A.O.A.C. chick unit.

Biologically tested fish oils of guaranteed vitamin D potency are the only oils that are suitable for supplying turkeys with vitamin D. Fish oils that have no guarantee of potency may or may not contain adequate vitamin D.

It is a good practice to add a chick-tested fish oil to the ration of the breeding flock to insure an adequate storage in the egg for development of the embryo and for starting the poult after it is hatched. Since sunshine cannot be depended upon during the brooding season, fish oil should be a regular part of the ration until the poults are put on the range. Whether or not fish oil should then be continued depends on the amount of sunshine available. A proper balance of minerals, especially calcium and phosphorus, is also essential in preventing rickets.
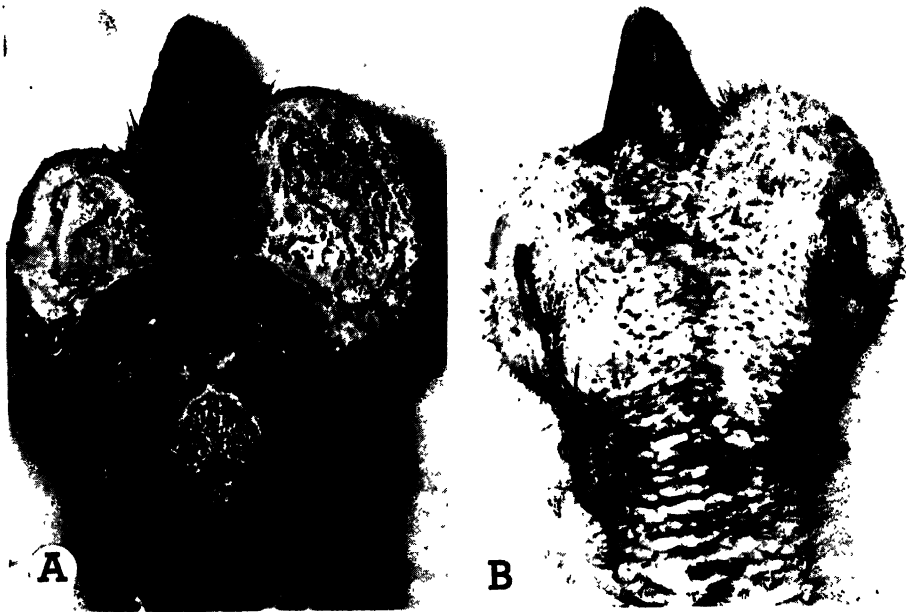


FIG. 40.5. An extreme case of sinusitis in a turkey hen suffering from vitamin A deficiency after being fed for 8 months on a ration containing a low level of vitamin A. The picture on the left (A) is a sagittal section of the head shown on the right. (B) Note the massive accumulation of the whitish-yellow caseous exudate typical of sinusitis as associated with vitamin A deficiency in turkeys. (Hinshaw, Univ. of Calif.)

## DIETARY DERMATITIS

Jukes (1938) showed that riboflavin-deficient diets caused symptoms of dermatitis in young poults. Patrick, Boucher, Dutcher, and Knandel (1941, 1943) were unable to verify Jukes' results but found that biotin is an anti-dermatitis factor for turkeys. Recently, using a purified basal diet, Jukes, Stokstad, and Belt (1947) were able to produce dermatitis identical to that formerly produced by Jukes when riboflavin was withheld. It would seem, therefore, that both riboflavin and biotin may be necessary to prevent dietary
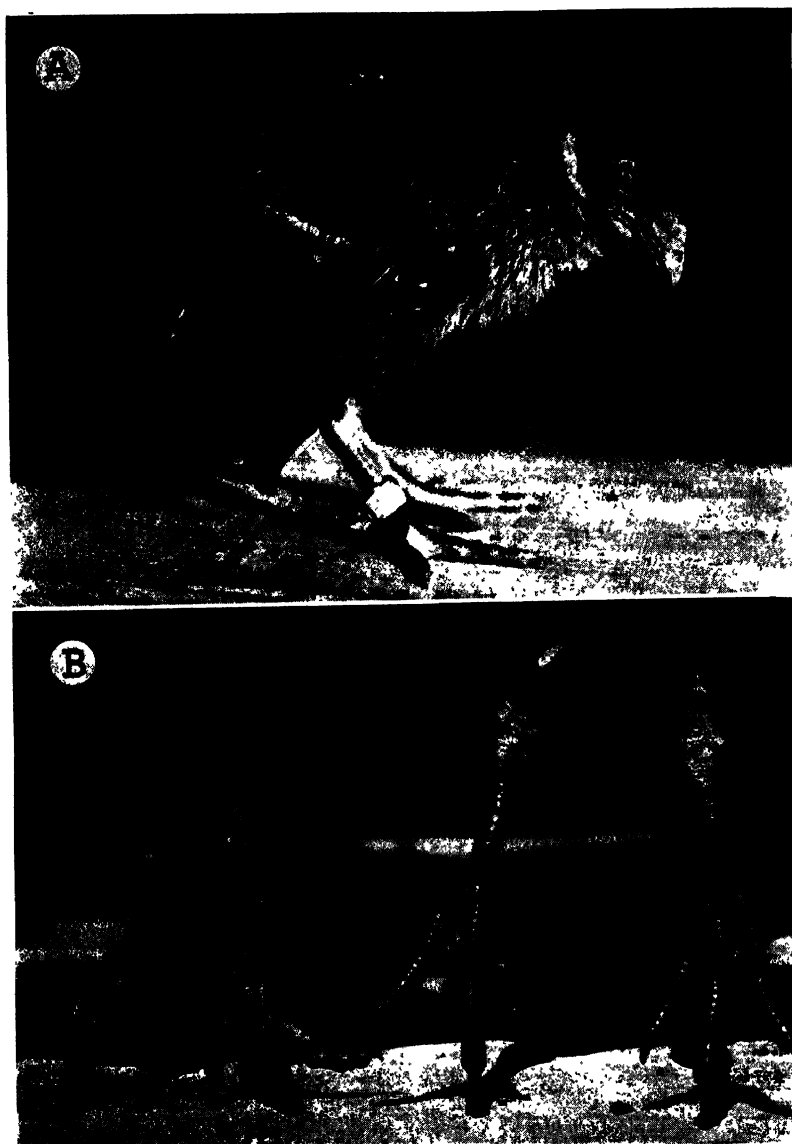
FIG. 40.6. A—29-day-old turkey after 17 days on a riboflavin-deficient diet. Note the encrusted eyelids, mouth, and nostrils. The feet did not show lesions at the time this picture was taken. B—legs and feet of two three-week-old turkeys. The ones on the left are from a poult fed a flavin-deficient ration from hatching time. The others are from a poult fed the same basal ration, with flavin added. Note the dryness of the skin of the legs and the marked ulceration of the foot pads in the affected specimen. (T. H. Jukes.)

dermatitis. Niacin deficiency (Kratzer, 1947) may also cause inflammation of the mouth, which must be differentiated.

This deficiency disease may be seen in the field, but other forms of derma-

titis are also common and must be considered in making a diagnosis. Very little is known regarding the other types of dermatitis (see miscellaneous diseases).

**Symptoms.** The symptoms of dietary dermatitis in poults consist of a sore mouth and encrustations at the corners of the mouth; diarrhea, resulting in
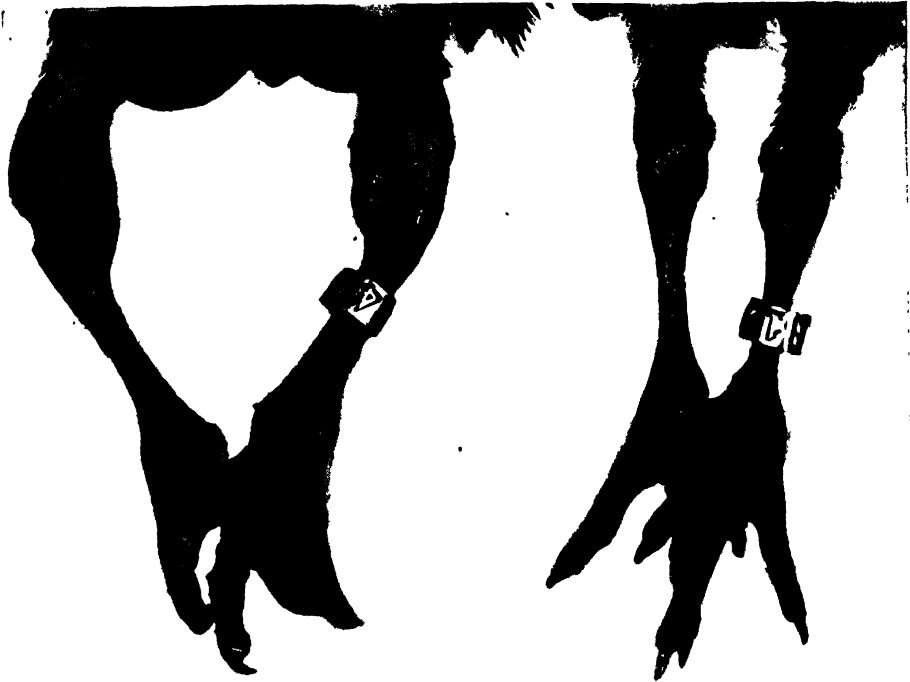


Fig. 40.7. Perosis. Note thickening, shortening, and distortion of the perotic legs on the left as compared with the normal legs on the right. (T. H. Jukes, Jour. of Nutr.)

an inflammed encrusted vent; thickened eyelids that tend to stick together; ragged feathers; and a listless, unthrifty appearance. In advanced cases the feet may also be involved (Fig. 40.6 A and B). Growth is very slow, and mortality is high.

Pantothenic acid (filtrate factor) is sometimes spoken of as the "chick antidermatitis vitamin." It appears not to prevent dermatitis in poults, but if they are placed on a diet deficient in this vitamin they grow slowly, and many of them die in a short time. This substance is found in moderate amounts in most poultry feeds.

**Prevention and control.** Riboflavin is very important for the promotion of growth and the production of hatchable eggs. Fresh greens and alfalfa meal are the cheapest source of riboflavin, which is also present in milk or whey. Poult starting mashes should contain alfalfa meal and, in addition, dried

milk or dried whey as sources of riboflavin. Older turkeys may receive their complete riboflavin requirement from fresh greens and alfalfa meal. (See Table 1 for other sources of riboflavin and for sources of biotin and niacin.)



FIG. 40.8. Leg bones (2, 4, 6) from a perotic four-week-old turkey poult receiving a ration deficient in choline, compared with leg bones (1, 3, 5) from a normal poult. Note the shortening, thickening, and distortion of the bones caused by choline deficiency. (T. H. Jukes, Jour. of Nutr.)

## PEROSIS

(Slipped Tendon, Hock Disease, Spraddle Legs)

Perosis (Figs. 40.7, 40.8, and 40.9) may cause considerable loss to turkey growers if not prevented by use of a properly balanced ration. Jukes (1940) and Evans, Rhian, and Draper (1943) have shown that this condition in turkeys is associated with an improper balance of calcium, phosphorus, manganese, and choline in the ration. According to Patrick et al. (1943) biotin is also an antiperosis factor, and according to Briggs (1946) and Jukes et al. (1947) niacin is necessary to prevent it.

This disease should not be confused with a similar condition of newly hatched poults which is also called "spraddle legs." This latter condition is caused by one of a number of factors including faulty incubation, improper diet in breeding stock, and faulty structure of hatching trays.

Symptoms. The symptoms seen in this disease are bowed, or badly twisted legs due to improper calcification of the tibia and metatarsus, especially at the hock joints. This deformity allows the tendon of Achilles to slip from

its groove. In turkeys the meta-
tarsus often turns at a right
angle giving the name "sprad-
dle legs" to the condition.
Occasionally, the femorotibial
joint is affected. There is
usually enlargement and flat-
tening of the hock joint, and
sometimes the entire shank.

**Prevention.** There is no
cure for the disease after it
reaches the deformity stages.
According to Asmundson and
Jukes (1939), a poult ration
containing from 0.8 to 1.0 per
cent phosphorus and 1.8 to 2.0
per cent calcium may be relied
upon to provide enough calcium
and phosphorus for bone forma-
tion and at the same time pre-
vent perosis under ordinary
conditions.

Hopper feeding of limestone
grit to growing turkeys is not
recommended as this practice
is unnecessary and dangerous
since it upsets the mineral



FIG. 40.9. An advanced case of slipped tendon
in a mature turkey. Note the rotation of the
right leg at the hock joint. (Hinshaw, Univ. of
Calif.)

balance of the ration. If grit is to be supplied, it should be of an insoluble
type. A summary of the sources of the vitamins needed to prevent perosis,
and the amounts needed are given in Table 1.

REFERENCES

Asmundson, V. S., and Jukes, T. H.: 1939. Turkey production in California. Calif. Agr. Ext.
    Circ. 110.

Briggs, G. M.: 1946. Nicotinic acid deficiency in poults and the occurrence of perosis. Jour.
    Nutr. 31:79.

Evans, R. J., Rhian, M., and Draper, C. I.: 1943. Perosis in turkey poults and the choline
    content of their diets. Poultry Sci. 22:88.

Ewing, W. R.: 1943. Handbook of Poultry Nutrition. W. R. Ewing Publishing Co., So. Pasadena,
    Calif.

Hinshaw, W. R., and Lloyd, W. E.: 1934. Vitamin A deficiency in turkeys. Hilgardia (Univ. of
    Calif.) 8:281.

Jukes, T. H.: 1938. The vitamin G requirements of young poults. Poultry Sci. 17:227.

———: 1940. Effect of choline and other supplements on perosis. Jour. Nutr. 20:445.

———, Stokstad, E. L. R., and Belt, M.: 1947. Deficiencies of certain vitamins as studied with
    turkey poults on a purified diet. I. Pteroylglutamic acid, riboflavin, niacin, and inositol. Jour.
    Nutr. 33:1.

Kratzer, F. H.: 1947. Vitamin requirements of turkeys. Turkey World 22:60.

Marsden, S. J., and Martin, J. H.: 1944. Turkey Management. The Interstate Publishers, Danville, Ill.

Patrick, H., Boucher, R. V., Dutcher, R. A., and Knandel, H. C.: 1941. Biotin and prevention of dermatitis in turkey poults. Proc. Soc. Exper. Biol. and Med. 48:456.

——, Boucher, R. V., Dutcher, R. A., and Knandel, H. C.: 1943. Prevention of perosis and dermatitis in turkey poults. Jour. Nutr. 26:197.

Scott, H. M.: 1937. Turkey production in Kansas. Kan. Agr. Exper. Sta., Bul. 276.

——, Hughes, J. S., and Loy, H. W.: 1932. Rickets in young turkeys. Poultry Sci. 11:177.

Wilgus, H. S.: 1940. Experiments show turkey poults need four times as much vitamin A as do chicks. Colo. Agr. Exper. Sta., Bul. 2:3.

# FUNGUS DISEASES

Fungus diseases, due to molds and yeasts, may cause considerable mortality in turkeys. The most important are aspergillosis, favus, and moniliasis.

## ASPERGILLOSIS
### (Brooder Pneumonia, Mycotic Pneumonia, Pneumomycosis)

Aspergillosis, more commonly known in the field as brooder pneumonia, is caused principally by *Aspergillus fumigatus,* although some other molds of the same genus may be responsible. *A. fumigatus* is widely distributed in nature and is pathogenic for many animals, including man. In young poults kept on contaminated litter, it produces pneumonia with heavy mortality. Infected older birds may suffer from pneumonia or air sac infection. Baker, Courtenay-Dunn, and Wright (1934) are of the opinion that molds belonging to other genera may also be responsible for mycotic pneumonia in birds.

Aspergillosis has been reported in turkeys by Lignières and Petit (1898), Balfour (1911), Schlegel (1915), Hinshaw (1937), and Durant and Tucker (1935). The outbreak reported by Durant and Tucker occurred in wild turkey poults reared in captivity. The disease appeared at 5 days of age and reached a maximum mortality at 15 days. When the epizootic subsided at the end of three weeks, only 200 of the 785 poults remained alive. Van Heelsbergen (1929) gives a detailed description of an outbreak investigated by Schlegel. The description given below taken from Hinshaw (1937) is essentially the same as described by Schlegel and is based on the writer's experience with the disease.

**Symptoms.** The symptoms depend on the seat of infection. Lesions in the mouth, trachea, or bronchi produce hoarseness, heavy breathing, and sometimes rattling in the throat. Birds suffering from air sac infection alone may not show any symptoms. As the disease progresses, dullness, labored breathing, and emaciation may be seen. Death probably results from either toxemia or asphyxiation. The mortality varies but is usually greater in brooder poults than in older birds.

**Autopsy findings.** Diagnosis is readily made in advanced cases. The lungs and air sacs are the principal seats of infection, but the process may extend into the peritoneal cavity or into the air passages of the bones (Fig. 40.10). The kidneys, liver, and spleen may be affected by direct contact from the

air sacs. Yellow, semiliquid, or caseated masses in the air sacs and lungs, with button-like ulcers attached to the mucous membranes, are common. In the early stages these ulcers appear as round, yellowish-white masses attached to the membrane. In advanced cases a greenish mold turf may be seen over the surfaces of the infected areas and in the convex depressions of the ulcers, especially in the air sacs. Final diagnosis depends on identification of the mold. The fungus can be readily demonstrated by microscopic examination



FIG. 40.10. Lungs of turkey showing typical caseous nodules seen in the early stages of aspergillosis. Note also irregular lesion with center darkened by aerial hyphae of the fungus, denoted by the arrow. (Hinshaw, Univ. of Calif.)

of specimens that have been treated with 10 per cent sodium or potassium hydroxide and by culturing on suitable media. A careful examination of the surface of the button-like lesions will often reveal aerial hyphae, and seedings from these will usually insure a pure culture.

**Prevention, control, and treatment.** Careful selection of grain and litter is essential in preventing this disease. Access to musty, moldy strawstacks should be avoided. A common source of the infection is semisolid milk from barrels that have been improperly cared for. If used, semisolid milk should always be kept covered with water to prevent mold growth over its surface.

Improperly kept drinking fountains used for dispensing milk have also been found to be a source of infection. One outbreak observed by the writer was associated with contaminated milk cans. The inside of the lids of the cans used for transporting milk was found to be covered with a fine mold growth; the owner had washed and scalded the cans daily, but thought it unnecessary to clean the lids. The storage barrels for the milk were also heavily infected.

The areas around feed hoppers and watering places are fertile fields

for the growth of molds. Unless a permanent yard system is used, frequent moving of feed troughs and watering places is advisable. Placing feed containers and watering fountains on screened elevated platforms helps to prevent turkeys from picking up molds that develop in such places. Drainage is advisable with areas where water is liable to stand after rains.

Control is best accomplished by removing the cause. A careful search should be made for mold in the litter, the feed, and the feed and water containers. Daily cleaning and disinfection of feed and water utensils with



Fig. 40.11. Favus. An unusual case affecting the entire carcass. (L. D. Bushnell.)

0.5 per cent copper sulfate solution will aid in eliminating the infection. Spraying of the ground around the containers with chemical solutions may be advisable if it is impossible to change feeding areas frequently. In outbreaks, a 1:2,000 solution of copper sulfate in place of all drinking water may be used to aid in preventing the spread through this means, though it should not be relied upon as a preventive to be used continually.

The deep-seated nature of the disease renders treatment of little avail. Extreme care should be used in handling and disposing of sick birds, because of the possible danger of transmitting the disease to the attendant.

### FAVUS

Favus is a chronic skin disease caused by a fungus, *Achorion gallinae*, and characterized by whitish areas on the exposed skin parts of the body (Fig. 40.11). It is not a commonly occurring disease. Since man is susceptible, care should be taken to prevent transmission if an outbreak occurs. The disease is generally mild and sporadic in nature. It may exist in a flock for several months, but few losses directly traceable to it are experienced.

**Symptoms.** The white powdery-like spots which characterize the disease

usually appear first around the beak. Thence they spread to the wattles, dewlap, and snood, and in extreme cases to the feathered portions. The fine pinpoint white spots finally coalesce and may cover a considerable area. As the fungus spreads and grows, a piling up of the threads occurs, and a thick, crustlike area may result.

**Prevention, control, and treatment.** Removal and disposal of all infected birds is recommended. It is well to move the flock to new quarters when practicable. After removal of infected individuals, the premises must be thoroughly cleaned and disinfected.

Treatment should be attempted only in very valuable birds. A mixture of 6 parts of glycerine and 1 part of iodine applied locally is recommended by van Heelsbergen (1929) for the infected parts of the head. Beach and Halpin (1918) reported that a formalin-vaseline ointment rubbed thoroughly into the lesions cured fifty out of fifty-two cases treated. This ointment may be prepared by thoroughly mixing and shaking in a closed jar 5 per cent by weight of commercial formalin, and melted vaseline. After mixing thoroughly the ointment is allowed to harden.

<div align="center">

MONILIASIS

(Mycosis of the Crop, Thrush)

</div>

Moniliasis is a disease of the upper digestive tract of both chickens and turkeys caused by yeastlike organisms belonging to the genus Monilia. Jungherr (1933a) was probably the first in the United States to observe the disease in chicks; Gierke (1932) has reported an outbreak of a thrushlike disease occurring in turkeys in California during the summer of 1932; and Hinshaw (1933) has described the results of studies on several outbreaks in turkeys and chickens. Wickerham and Rettger (1939) in a taxonomic study of Monilia species from various sources included several strains isolated by Hinshaw from turkeys and chickens. These proved to be *M. albicans* and indistinguishable from species isolated from man. Jungherr (1933b, 1934) found in his later studies that *M. albicans* and *M. krusei* and *Oidium pullorum* n.sp. were the most frequently isolated yeastlike fungi from chicks. Of these he considered *M. albicans* and *O. pullorum* of etiological importance.

Jungherr (1933a) was able to transmit the disease in chicks by feeding fecal material from diseased chicks and by injecting pure cultures of *M. albicans*. The average period of incubation under experimental conditions was 31 days. Hinshaw (1933) was able to transmit the disease from turkeys to turkeys, chickens, and rabbits. Jungherr also succeeded in isolating Monilia from the cloaca of laying hens affected with a moist type of vent gleet. He presented evidence that the causative organisms may live on the shell of the egg and carry the infection to young chicks in the incubator. He has not, however, definitely proved transmission by this means. Hinshaw

observed that the disease is one associated with poor management, where unsanitary surroundings prevail, and debilitation is prevalent. It must be differentiated from the disease formerly described by Jungherr (1927) as a mycosis of the crop of turkeys but now known to be caused by a Trichomonas. (See Trichomoniasis of Upper Digestive Tract.)

**Symptoms.** As most of the outbreaks observed have been complicated with some other pathologic condition, specific symptoms have been difficult to determine. More or less constant symptoms, however, are listlessness, loss of appetite, tendency to stand around with heads drawn back on the
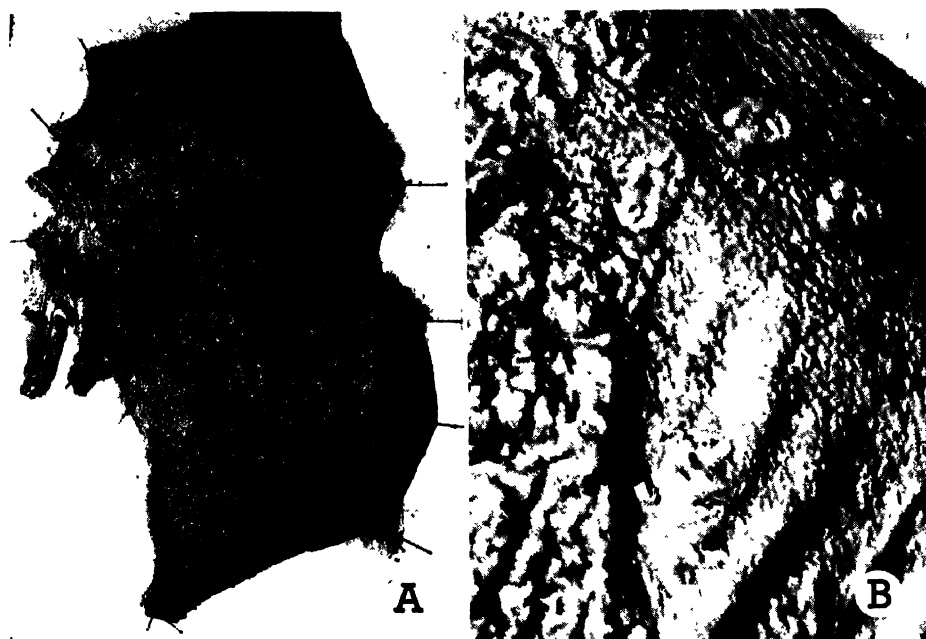


FIG. 40.12. A—crop of a turkey suffering from moniliasis. B—enlarged section of A; note the raised, piled-up exudate which tends to form roselike masses. (Hinshaw, Univ. of Calif.)

shoulders, and a sunken appearance of the chest. The eyes and sinuses appear sunken, and the heads haggard.

**Autopsy findings.** The crop has been the most common seat of infection. Fungi have also been demonstrated in scrapings from the mouth, infraorbital sinuses, upper and lower esophagus, proventriculus, gizzard, and intestines. Cultures of the causative organism have been obtained from all of these organs and in addition from a lung abscess and from a skin abscess.

In the more acute cases, as well as in the milder cases, there is seen a catarrhal to thick mucoid exudate with a tendency to form a pseudo-membrane. Soft, raised, whitish-yellow ulcers having a roselike appearance and scattered over the surface, at times coalescing to form a solid mass of

piled-up exudate (Fig. 40.12), characterize the more chronic cases. These lesions have been variously described by turkey growers and others as having a "turkish-towel-like" or "curdy" appearance. In early or mild cases the mucous membrane may appear parboiled. The lesions are easily scraped from the surface, leaving the mucous membrane abraded and injured to a greater or less extent, according to the severity of the infection.

In most cases the crops either are empty or contain a small amount of thick slimy exudate. Cultures of the fungus are readily obtained in nearly pure state by washing off the surface exudate and planting a fairly deep scraping on 2 per cent dextrose agar, glycerine dextrose agar, or on Sabouraud's honey agar of a pH of 6.0 to 6.4. The colonies are large enough and so characteristic that unless molds interfere, no difficulty is experienced in selecting individual colonies for purification.

**Prevention, control, and treatment.** Because of the nature of the disease and its frequent association with other diseases common in crowded quarters and in flocks suffering from some form of malnutrition, sanitation, and proper diet are important factors in control. Removal of birds to clean and thoroughly disinfected quarters, together with the daily cleaning and disinfection of feed and water containers, has helped to reduce losses.

Copper sulfate solution of a 1:2,000 dilution substituted for all drinking water for a few days is a helpful control procedure. It should be remembered, however, that turkeys do not like this solution; if any other source of water is available, they will not touch it. Use of crockery or wooden fountains is recommended when copper sulfate solutions are used. A convenient method of making up approximately a 1:2,000 dilution of copper sulfate solution is given under disinfectants on pages 107 and 108.

## REFERENCES

Baker, A. Z., Courtenay-Dunn, J., and Wright, M. O.: 1934. Observations on fungal pneumonia in the domestic fowl. Vet. Jour. (Brit.) 90:385.

Balfour, A.: 1911. Aspergillary pneumokoniosis in the lung of a turkey. Fourth Rep. Wellcome Res. Lab. (Gordon Mem. Coll.), Vol. A. (Med.) :353.

Beach, B. A., and Halpin, J. G.: 1918. Observations on an outbreak of favus. Jour. Agr. Res. 15:415.

Durant, A. J., and Tucker, C. M.: 1935. Aspergillosis of wild turkeys reared in captivity. Jour. Am. Vet. Med. Assn. 86:781.

Gierke, A. G.: 1932. A preliminary report on a mycosis of turkeys. Calif. St. Dept. Agr., Monthly Bul. 21:229.

Hinshaw, W. R.: 1933. Moniliasis (thrush) in turkeys and chickens. Proc. Fifth World's Poultry Cong., Paper 97:1.

————: 1937. Diseases of turkeys. Calif. Agr. Exper. Sta., Bul. 613.

Jungherr, E.: 1927. Two interesting turkey diseases. Jour. Am. Vet. Med. Assn. 71:636.

————: 1933a. Observations on a severe outbreak of mycosis in chicks. Jour. Agr. Res. 46:169.

————: 1933b. Studies on yeast-like fungi from gallinaceous birds. Storrs Agr. Exper. Sta., Bul. 188.

————: 1934. Mycosis in fowl caused by yeast-like fungi. Jour. Am. Vet. Med. Assn. 84:500.

Lignières and Petit: 1898. Péritonite aspergillaire des dindons. Rec. Méd. Vét. 75:145.

Schlegel: 1915. (Quoted by van Heelsbergen.)

van Heelsbergen, T.: 1929. Handbuch der Geflügelkrankheiten und der Geflügelzucht. Ferd-
    inand Enke, Stuttgart, 312.
Wickerham, L. J., and Rettger, L. F.: 1939. A taxonomic study of *Monilia albicans* with special
    emphasis on morphology and morphological variation. Jour. Trop. Med. and Hyg. 42:174,
    187, and 204.

## BACTERIAL AND VIRUS DISEASES

The common diseases of turkeys caused by bacterial and virus agents are included in this section. The reader is referred to standard textbooks on bacteriology if descriptions of the causative organisms are desired. For discussions on fowl pest, fowl coryza, equine encephalomyelitis virus in birds, Newcastle disease, and other diseases seen in turkeys, the reader is referred to the section on diseases of chickens.

### BOTULISM

Botulism is caused by toxin produced by an anaerobe, *Clostridium botulinum*. Of the three types of botulinus toxins poisonous to man and animals, only A and C are known to affect fowls. The toxins are produced by the microorganism while growing in such substances as decomposing food, dead carcasses, and wet grain, and are transmitted to birds when the contaminated products are eaten. Coburn and Quortrup (1938) have described an outbreak of botulism in turkeys caused by the type C organism. The outbreak occurred in a flock of 1,400 turkeys which were ranging on a 20-acre stubble field. About fifty turkeys were sick at the time of the investigation, and an additional fifty had died the previous week. The source of the toxin was found to be a shallow, stagnant pool of water in the stubble field. Filtered water samples from it were shown to contain the type C toxin by tests on white mice. *Clostridium botulinum* (type C) was also isolated from the soil taken from the water hole.



FIG. 40.13. Typical posture in botulism of turkeys. (Hinshaw, Univ. of Calif.)

**Symptoms.** The most common symptom is complete paralysis of the neck, which gives the disease its name, "limberneck." The birds sit with their heads and necks on the ground or extended over the back (Fig. 40.13), often in a comatose condition. In turkeys the feathers do not shed so readily as in chickens affected with the disease.

The turkeys in the outbreaks described by Coburn and Quortrup (1938) manifested evidence of cyanosis of the head, posterior paralysis, and dyspnea, but only a few showed paralysis of the nictitating membrane, a symptom

usually considered pathognomonic. Some of the sick turkeys recovered spontaneously.

**Autopsy findings.** Coburn and Quortrup described the following post-mortem findings: petechial hemorrhages on the auricular pericardium, hyperemia of the duodenal mucosa, and cloaca distended with urates. Thus the gross pathology would make one suspicious of fowl cholera.

One should look for evidence of spoiled food in the crop and for the presence of fly maggots which are suggestive of the consumption of spoiled food. Diagnosis depends on the history obtained and on the symptoms and autopsy findings, but finally on the demonstration of the toxin or causative organism.

**Prevention, control, and treatment.** Every effort should be made to prevent turkeys from obtaining foods that might harbor the botulinus organism. Spoiled canned vegetables should never be given, for they are liable to contain botulinus toxin.

When the disease appears, all the birds should be moved to a new feeding ground and, if necessary, fenced in to prevent access to spoiled food. Sick birds should have plenty of shade. Their crops can be drained and flushed out with warm water with the aid of a rubber tube and a funnel or by the method shown in Figure 40.51. Large doses of mineral oil or castor oil may help to get rid of the toxin in birds that have not gone into coma. The cause of the trouble should be traced, and recurrence prevented. In valuable birds polyvalent (mixed) botulinus antitoxin may be used.

Persons handling turkeys suffering from botulism should keep in mind that the botulinus toxin may affect man. Careful washing of the hands after care of the birds is suggested as a precautionary measure.

REFERENCE

Coburn, D. R., and Quortrup, E. R.: 1938. Atypical botulism in turkeys. Jour. Am. Vet. Med. Assn. 93:385.

## ERYSIPELAS

This disease caused by the swine erysipelas organism, *Erysipelothrix rhusiopathiae,* was first reported in turkeys by Jarosch (1905). The first report of an outbreak in the United States was made by Beaudette and Hudson (1936) in New Jersey. A complete bibliography of the literature to that date is included in their paper. Since 1936 it has been diagnosed in various sections of the United States. Published reports from the following states have been made: Utah by Madsen (1937); New York, Vermont, and Massachusetts by Van Roekel, Bullis, and Clarke (1938) and Brunett and Hofstad (1943); California by Hoffman and Hinshaw (1938); Oregon by Rosenwald and Dickinson (1939, 1941); Minnesota by Schlotthauer and Thompson (1940); Washington by Lindenmayer and Hamilton (1942)

and Lindenmayer (1943); Connecticut by Jungherr and Gifford (1944); and Colorado by Stiles (1946).

Most of the outbreaks reported by American investigators have been confined to single flocks, and recurrence of the disease has seldom been reported from the same ranch. Sheep rather than swine have been the most probable factors in transmission of the disease in turkeys in the United States.

The outbreaks have usually occurred in turkeys approaching the market age, and males have appeared to suffer the heaviest losses. Rosenwald and



FIG. 40.14. Erysipelas. Swollen snood which is, according to Rosenwald and Dickinson, pathognomonic for the disease may also be seen in outbreaks of fowl cholera. (Rosenwald and Dickinson, Am. Jour. Vet. Research.)

Dickinson (1941) have, however, diagnosed the disease in poults from a few weeks of age to maturity.

**Symptoms and mortality.** The disease manifestations as described by Beaudette and Hudson (1936) are primarily those of a septicemia. The mortality in the outbreak studied by them was high, 200 of a flock of 500 dying in 9 days. This is a heavier mortality than that reported by most investigators.

The symptoms are listlessness, drooping tails and wings, and sometimes yellowish-green diarrhea. Swelling of the joints of the legs have been noted, but this is not a constant symptom. Madsen reported that the affected birds remained aloof from the remainder of the flock. This was also noted by Hinshaw. These sick birds have a tendency to crouch, the heads often appear cyanotic, and nasal catarrh is a common symptom. Swelling of the snood, as illustrated in Figure 40.14, is characteristic of this disease, but similar swellings are also common in outbreaks of fowl cholera. Erysipeloid lesions

may appear on the face, involving the major portion of the eyelids and area posterior to them (Fig. 40.15). The effect on the appetite seems to vary in different outbreaks, though most investigators agree that feed consumption is lowered. Both acute and subacute forms are seen in turkeys. Recovery in some acute cases, according to Van Roekel, Bullis, and Clarke (1938), may be complete in a week or 10 days.

The effect of the disease on body temperature of birds is not reported in the available literature. According to Van Es and McGrath (1936), infected



Fig. 40.15. Erysipelas. Erysipeloid lesion on the face of a turkey, involving the major portion of eyelids and an area posterior to them. (Ore. Agr. Exper. Sta.)

swine may show temperatures as high as 110° F. in acute outbreaks. Hinshaw observed temperatures in field cases of sick turkeys as high as 109.6° F., but rise in temperature has not been consistent with symptoms.

Adult turkey hens artificially infected with *E. rhusiopathiae* have not shown marked increase in temperature. One such bird (nineteen weeks of age) given 0.8 cc. of 24-hour broth culture became ill in 48 hours, and showed a continued rise in temperature until it reached 110.6° F. on the fourth day after inoculation. On the day of its death (the sixth after inoculation) the temperature was 109.8° F. Another bird which became visibly ill in 48 hours, but recovered within two weeks, showed a very slight rise in temperature during the period of visible symptoms. A third turkey hen showed an increase from 104.9° F. on the day before inoculation to 108.8° F. on the sixth day after inoculation. Its temperature gradually subsided, though the bird itself developed a chronic type of the disease and was finally killed in an emaciated condition in four weeks.

**Autopsy findings.** Diffuse hemorrhagic areas of various sizes are common in the breast muscles. The skin of the breast may appear pink, but no diamond-shaped lesions such as are seen in swine have been described. The nasal passages are usually filled with thick mucus; the livers are enlarged, congested, and friable. Catarrhal enteritis is evident, with some reddening of

the mucosa of the large intestine. In most cases the spleens are enlarged, mottled, and friable, hemorrhages sometimes appearing. Other lesions occasionally found are hemorrhages in the pericardium, congestion of the kidneys and lungs, and rarely, browning of the lung tissue. Stiles (1946) considered the absence of pus in affected joints and other structures as characteristic of the disease.

**Diagnosis.** The marked hemorrhagic condition of the fascial and muscular tissues of the breast is the most significant finding at autopsy according to most investigators. Diagnosis must be confirmed by isolation of the causative organism. Differentiation from fowl cholera is necessary because of the similarity of the two diseases. The use of mice and pigeons for inoculation tests is recommended, as is the use of anti-swine erysipelas serum for neutralization tests in the inoculated test animals. Mice or pigeons given either the pure cultures of the organism or tissues from erysipelas cases die within 24 to 96 hours. Similarly infected animals protected with 0.5 cc. of antiserum do not become sick.

Another aid to diagnosis is to stain blood or liver smears by Gram's method. The characteristic Gram-positive rods (which decolorize easily, however) are usually grouped in interlacing bundles. The individual organisms are slender, slightly curved, and show characteristic granules. It is possible to isolate the causative organism from the bone marrow of turkeys which have been dead as long as two weeks. Hinshaw has even isolated it from the marrow of the small metatarsal bones removed from carcasses that have lain on the range for at least two weeks.

**Prevention, control, and treatment.** Since the disease is common in swine and sheep in the United States, turkeys should be kept away from swine and sheep herds, at least in areas where erysipelas is known to exist. Removal of the infected flock to a new range and segregation of the sick birds is recommended. The temperatures of the birds may be an aid in determining the sick birds but cannot be depended upon to detect all of the birds in the early stages. The use of anti-swine erysipelas serum in sick flocks has been suggested, but inconsistent results have been reported. Van Roekel, Bullis, and Clarke (1938) reported favorable results in one outbreak where ninety-one exposed turkeys were given 10 cc. of swine erysipelas antiserum to each bird by the intraperitoneal route. In contrast, Rosenwald and Dickinson (1941) state that satisfactory results were not obtained by its use under controlled field and laboratory conditions.

Beaudette and Hudson (1936) found that serum from recovered turkeys protected mice against infection. Lindenmayer and Hamilton (1942) injected 3.0 cc. of formalized serum from sick turkeys intramuscularly and obtained protection and curative action in exposed turkeys. They used only a few turkeys in their trials, and as far as is known, no one has confirmed their results.

Gifford and Jungherr (1946) reported successful results on the use of penicillin for control of the disease in turkeys. They obtained complete recoveries of sick turkeys which received two intramuscular injections of 25,000 Oxford units in 2 cc. of saline at 8-hour intervals. Grey (1947a) reports good results using four daily injections of 20,000 units each. He used the drug suspended in peanut oil and injected it into the wattles.

Grenci (1943) isolated *E. rhusiopathiae* from two samples of fish meal, a common ingredient of turkey feed. Therefore, this product should be thoroughly sterilized before being used in turkey feed, as a preventive measure.

Debeaking the males in a flock at the outset of the disease in order to prevent spread by fighting is suggested by Lindenmayer (1943). Reports from the field indicate that this procedure is worthy of trial. An electrically operated debeaking apparatus is available for this type of operation (see cannibalism).

McCulloch and Fuller (1941) found that household lye (sodium hydroxide) in dilutions of 1:200 to 1:500 is an effective disinfectant against *E. rhusiopathiae*. Phenol, liquor cresolis and related disinfectants, tincture of iodine, triethanol-ammoniumlauryl sulfate, and household soaps were moderately effective, but formalin was ineffective.

**Caution.** Since the causative organism of this disease is pathogenic for man, extreme care should be taken when handling infected birds or tissues. Erysipeloid cases caused by contact with diseased turkeys have been reported by Stiles (1946). At least three additional cases (unpublished) have been reported to Hinshaw.

Grey (1947b) reports that a single 1 cc. dose of 140,000 micrograms of streptomycin was 100 per cent effective against artificially produced cases of the disease. One-half that dosage was only 70 per cent effective.

## REFERENCES

Beaudette, F. R., and Hudson, C. B.: 1936. An outbreak of acute swine erysipelas infection in turkeys. Jour. Am. Vet. Med. Assn. 88:475.

Brunett, E. L., and Hofstad, M. S.: 1943. Erysipelas in turkeys in New York State. Cornell Vet. 33:105.

Gifford, R., and Jungherr, E.: 1946. A report on penicillin treatment of swine erysipelas in turkeys. Mich. St. Vet. 7:18–19, 40–41.

Grenci, C. M.: 1943. The isolation of *Erysipelothrix rhusiopathiae*, and experimental infection of turkeys. Cornell Vet. 33:56.

Grey, C. G.: 1947a. Penicillin in the treatment of *Erysipelothrix rhusiopathiae*-infected turkeys. Vet. Med. 42:177

———: 1947b. Streptomycin in the treatment of *Erysipelothrix rhusiopathiae*-infected turkeys. Vet. Med. 42:216.

Hoffman, H. A., and Hinshaw, W. R.: 1938. Erysipelas of turkeys. Poultry Sci. 17:443.

Jarosch, L. W.: 1905. Über die Septikämie der Truthühner. Oesterr. Monatschr. für Tierheilk. 30:197.

Jungherr, E., and Gifford, R.: 1944. Three hitherto unreported turkey diseases in Connecticut: erysipelas, hexamitiasis, mycotic encephalomalacia. Cornell Vet. 34:214.

Lindenmayer, J. E.: 1943. Swine erysipelas in turkeys in the State of Washington. Jour. Am. Vet. Med. Assn. 102:368.

Lindenmayer, J. E., and Hamilton, C. M.: 1942. Treatment of swine erysipelas in turkeys with serum from a turkey infected with *Erysipelothrix rhusiopathiae*. Jour. Am. Vet. Med. Assn. 100:212.

Madsen, D. E.: 1937. An erysipelas outbreak in turkeys. Jour. Am. Vet. Med. Assn. 91:206.

McCulloch, E. C., and Fuller, S. A.: 1941. The relative efficiencies of disinfectants in killing *Erysipelothrix rhusiopathiae*. Am. Jour. Vet. Res. 2:77.

Rosenwald, A. S., and Dickinson, E. M.: 1939. A report of swine erysipelas in turkeys. Cornell Vet. 29:61.

———— and Dickinson, E. M.: 1941. Swine erysipelas in turkeys. Am. Jour. Vet. Res. 2:202.

Schlotthauer, C. F., and Thompson, L.: 1940. The occurrence of erysipelas in turkeys. Jour. Am. Vet. Med. Assn. 96:103.

Stiles, G. W.: 1946. Observation of swine erysipelas in turkeys. (Including the public health aspect and possible human cases.) Jour. Am. Vet. Med. Assn. 109:65.

Van Es, L., and McGrath, C. B.: 1936. Swine erysipelas. Nebr. Agr. Exper. Sta., Res. Bul. 84.

Van Roekel, H., Bullis, K. L., and Clarke, M. K.: 1938. Erysipelas outbreaks in turkey flocks. Jour. Am. Vet. Med. Assn. 92:403.

## FOWL CHOLERA

Fowl cholera, caused by *Pasteurella avicida* results in severe economic losses to turkey growers in certain areas. It was first described by DeVolt and Davis (1932), who reported an outbreak in a flock of 175 turkeys in Maryland where there was a 17 per cent mortality. Moynihan and Bankier (1945) have reported the disease in Canada, and Smith and Field (1944) described an outbreak in England. In the writer's experience, the disease has been most prevalent in turkeys of about marketable age (six to eight months). In many outbreaks, chickens have been shown to be the source of the disease, but recent observations indicate that it may also be carried by adult turkeys. The organisms isolated from turkeys are identical to those recovered from chickens (see fowl cholera in the general section for literature citations).

**Symptoms, course, and mortality.** In many respects the symptoms of fowl cholera resemble those seen in fowl typhoid outbreaks. They include increased thirst, loss of appetite, listlessness, yellow or greenish-yellow, watery diarrhea, and a rise of 2° to 3° F. above the normal temperature. The heads appear blue to purplish and have a haggard, drawn appearance. A slimy to gelatinous exudate in the mouth and nostrils is not uncommon. The breast muscles become congested, and the skin appears pinkish. Swelling of the snood, similar to that seen in outbreaks of erysipelas (Fig. 40.14) is common in males. Paralysis of the legs and swollen joints are also common symptoms noted in both males and females which develop the chronic type of the disease.

The course of the disease is acute, heavy losses occurring within a few days, followed by intermittent losses. Symptoms may not be observed before death. In less acute cases the birds may linger for several days before dying. Very few sick turkeys recover. Reports of losses of from a few birds to over half of the flock are common.

**Necropsy findings.** The necropsy findings in turkeys are typical of those in chickens, though generally more pronounced. The breast muscles are con-

gested, and the crop usually contains considerable food having a very sour odor. The heart is often enlarged, and the pericardium may be thickened and covered with a whitish-yellow exudate. Many pin-point hemorrhages (petechiae) are commonly found over the surface of the pericardial sac, the muscles of the heart, and the adjacent tissues. The pericardial sac may be filled with a yellowish fluid containing whitish-yellow flakes. The liver is never more than slightly enlarged. It is friable, often salmon colored, and may contain many minute whitish abscesses that give it a mottled appearance. The spleen may be enlarged or may show no alteration.

The blood vessels of the mesentery and intestines are usually engorged. The intestines lack tone and often show considerable evidence of hemorrhage, especially in the duodenum. The contents range from a semiliquid to a mucoid consistency. The feces are usually yellow to yellow-green.

The gizzard seldom contains much food, but the few contents present have a peculiar sour odor. In most cases the mucous membrane peels readily, and the muscle of the gizzard appears more red than normally. Considerable gelatinous exudate may be present in the proventriculus, and the mucous membrane may be denuded.

Pneumonia is a common finding in turkeys suffering from fowl cholera. Various stages of lung involvement from congestion to complete hepatization are seen. In such cases, the pleural cavity may contain a surplus of fluid, or the air sacs may be filled with a semisolid, yellow caseous mass. Similar caseous deposits may be found throughout the abdominal cavity, and such lesions must be differentiated from aspergillosis. In the latter condition, the characteristic nodules, and "button ulcers" serve as differentiating lesions.

There is a characteristic fetid odor to the body cavities and the digestive tract contents of birds suffering from fowl cholera. This odor, difficult to describe, is that of advanced putrefaction and is recognized after experience with a few cases. Isolation of the causative organism is the final means of diagnosis.

**Prevention, control, and treatment.** Sanitation and hygiene play an important role in prevention. Turkeys should be kept segregated from all other fowl that have suffered from the disease. Adult carriers are responsible for the yearly recurrence of the disease on some ranches, and depopulation for a season may be necessary. There is no evidence that the disease is transmitted through the egg, but it is not a good plan to keep for breeding purposes turkeys that have recently suffered from the disease. Complete segregation of the breeding and brooding units or sale of the entire adult flock before any poults are hatched are recommended as preventive measures on ranches where the disease has existed.

Skidmore's (1932) observations on the common housefly as a possible carrier emphasize the need for keeping turkeys well isolated from chickens or

other fowl that might be suffering from the disease, and for prompt destruction of all diseased birds. Wild birds must be considered possible transmitters of the disease. The type of Pasteurella which caused a recent outbreak studied at the California station (unpublished data) proved to be identical to the one isolated from quail by Hinshaw and Emlen (1943). Burning, or the use of a disposal pit, instead of burial of dead birds is recommended; otherwise the diseased carcasses, a source of infection, may be dug up by dogs and other animals. The general recommendations given on pages 112 to 114 are suggested as other precautions.

Certain of the sulfonamides have given promise as aids in controlling fowl cholera in chickens (Delaplane, 1945; Kiser, Greene, Prier, and Bottorff, 1947). Alberts and Graham (1948) employed sulfamerazine in an experimentally produced outbreak of fowl cholera in turkeys and in a naturally occurring outbreak in adult turkeys. Treated mash containing 0.5 per cent of the drug or 0.5 grain per pound of body weight given orally twice daily prevented losses from the disease during the period of treatment, but failed to prevent recurrence of the disease. Their findings suggested sufficient retention of sulfamerazine in the body after discontinuing treatment, to suppress *P. avicida* for 2 days after the course of therapy, or long enough for the owner to arrange for marketing the healthy birds. At the California station (unpublished data) sulfamethazine given at the rate of 0.3 per cent in the mash for a period of 3 days definitely reduced losses if given early in an outbreak. The drug did not, however, prevent a recurrence of the disease, nor were carriers eliminated. No evidence of immunity was noted following intermittent treatments over a period of several weeks, such as was reported by Kiser *et al.* (1947). Sulfamethazine in amounts greater than 0.3 per cent affected egg quality in laying flocks of turkeys. Penicillin was ineffective in trials made at the California station (unpublished data).

## REFERENCES

Alberts, J. O., and Graham, R.: 1948. Sulfamerazine in the treatment of fowl cholera in turkeys. Jour. Am. Vet. Med. Assn. In Press.

Delaplane, J. P.: 1945. Sulfaquinoxaline in preventing upper respiratory infection of chickens inoculated with infective field material containing *Pasteurella avicida*. Am Jour. Vet. Res. 6:207.

DeVolt, H. M., and Davis, C. R.: 1932. A cholera-like disease in turkeys. Cornell Vet. 22:78.

Hinshaw, W. R., and Emlen, J. T.: 1943. Pasteurellosis in California valley quail. Cornell Vet. 33:351.

Kiser, J. S., Greene, L. M., Prier, J., and Bottorff, C. A.: 1947. Treatment of experimental and naturally occurring fowl cholera with sulfamethazine. Poultry Sci. 26:546.

Moynihan, I. W., and Bankier, J. C.: 1945. An outbreak of fowl cholera in turkeys. Canad. Jour. Comp. Med. and Vet. Sci. 9:46.

Skidmore, L. V.: 1932. The transmission of fowl cholera to turkeys by the common housefly (*Musca domestica* Linn.) with brief notes on the viability of fowl cholera microorganisms. Cornell Vet. 22:281.

Smith, H. W., and Field, H. I.: 1944. The isolation of *Pasteurella aviseptica* from a turkey. Vet. Jour. 100:35.

## FOWL POX

Fowl pox, a disease of the unfeathered parts of the birds' bodies, is characterized by the formation of pustules and scablike processes. It is caused by a filtrable virus, pathogenic for chickens as well. Brunett (1933) tested fowl pox viruses of chicken, turkey, and pigeon origins and reported that the turkey strain was pathogenic for chickens, but not for pigeons. All three strains produced lesions in turkeys, but only the chicken and turkey strains produced immunity.

Tietz (1933) reported that turkeys were not susceptible to the strain of pigeon pox viruses used by him. Hinshaw's experience has been more like that of Tietz, though very slight swellings of the feather follicles have been noted. Coronel (1934), Brandly and Dunlap (1938), and Beaudette and Hudson (1941) published evidence to show that there is a distinct strain of turkey pox virus which differs from both the chicken and pigeon types. Field observations made by Hinshaw suggest that such is the case, and that the use of vaccines of chicken pox origin produce only temporary immunity in such outbreaks.

Matheson, Brunett, and Brody (1931) reported transmission of fowl pox by mosquitoes, and later Brody (1936) published a comprehensive report on subsequent investigations. These investigators found that one species of mosquito (*Aedes aegypti*) was still able to transmit pox 41 days after an initial infective meal. Blanc



Fig. 40.16. Fowl pox in turkeys. Taken three weeks after lesions first appeared. (Hinshaw, Univ. of Calif.)

and Caminopetros (1930) in an earlier paper reported transmission by *Culex pipiens* 58 days after feeding on infected birds.

**Symptoms and autopsy findings.** The first indication of pox is the appearance of minute yellowish eruptions on the dewlap, snood, and other head parts. They are soft and, in this pustular stage, easily removed, leaving an inflamed area covered with a sticky serous exudate. The corners of the mouth, the eyelids, and the oral membranes are commonly affected. The lesions enlarge and become covered with a dry scab or a wartlike mass of yellowish-red or brown color (Figs. 40.16 and 40.17). The number of lesions depends on the virulence of the disease. In young poults, the head, legs, and

feet may be completely covered with pustules. The disease may even spread to the feathered parts of the body (Fig. 40.18).



FIG. 40.17. Fowl pox lesions in mouth and esophagus. (Hinshaw, Univ. of Calif.)

Brandly and Dunlap (1938) reported two cases in three-week-old poults in which the foot pads and foot webs were involved. Large wart-like processes developed which made it difficult for the poults to walk. Except for a lesion of the mouth in one poult the disease was confined to the feet (Fig. 40.19). In these instances the infection was apparently introduced when the owner "toe punched" the poults for identification purposes.

Males often suffer more than females from the disease, probably because of their inclination for fighting, which spreads the infection through small lacerations.

The mouth parts, the tongue, the esophagus, and occasionally the crop, may be covered with masses of soft, yellow cankers closely adhering to the mucous membranes (Figs. 40.17 and 40.20). These yellow, diphtheric ulcers of fowl pox must be differentiated from the small, deep-seated, irregular, diphtheric ulcers or cankers often seen in the mouths of turkeys and not associated with typical head lesions. These cankers are common in turkeys that have been vaccinated against fowl pox or that have recovered from an outbreak. Their cause is not known.

Frequently during the breeding season atypical cases of fowl pox appear in adult turkeys which have been vaccinated with chicken pox vaccine several months previous. In these outbreaks, which usually involve only a small percentage of the birds, the



FIG. 40.18. Fowl pox lesions on the skin of breast of an adult turkey hen. The head parts were also severely affected. (Hinshaw, Univ. of Calif.)

mucous membranes of the eyes and the mouth are the principle parts affected. Externally, no lesions in the eyes may be visible, but when the inner surfaces of the lids are examined soft yellow diphtheric ulcers will be found to be the

cause of the increased lacrimation and inflammation of the eye. Typical yellow cankers described above characterize the mouth lesions. There are no internal lesions that are characteristic of the disease.

**Course and mortality.** There is a marked difference in the severity of cases of fowl pox in turkeys and, consequently, variations in the course of the disease. Whereas mild cases may clear up in two or three weeks, severe outbreaks often last for six, seven, or even eight weeks. The canker or mouth types take longer to clear up. In such cases, starvation is the cause of death. Blindness often occurs, after the closing of the eyes by severe infection of the eyelids. When the eye is involved, a yellowish canker-like lesion develops on the mucous membrane of the lid.



Fig. 40.19. Fowl pox lesions in the foot pads accidentally introduced during toe marking operations. (Dept. of An. Path. and Hyg., Univ. of Ill.)

The flock mortality is usually low, most of the losses being caused by blindness or starvation. Setback in development and loss in weight are of greater financial importance in the growing flock than the loss by deaths. As outbreaks commonly occur a few days or weeks before market time, it is often necessary to postpone killing the birds for several weeks. If the flock escapes an outbreak before market time, the disease sometimes appears in the breeding birds and causes severe losses through lowered egg production and poor fertility.

**Prevention.** Vaccination with live-virus vaccine, together with the usual sanitary program, is the recommended method for preventing fowl pox in turkeys. *The problem differs from that in chickens* because in the latter the effect of the disease and vaccination on egg production must be considered in the preventive program, while in the former a meat-producing bird only is involved. Furthermore, according to data collected on several thousand turkeys over a six-year period, healthy turkeys respond to vaccination, even when virus of chicken origin is used, with little or no systemic disturbance, such as sometimes follows vaccination of chickens. Vaccines made from turkey strains of virus cause more severe reactions than those made from chicken types, and the duration of immunity is no greater. Pigeon type vaccines produce little or no immunity in turkeys and should not be used.

**Need for vaccination.** Fowl pox is so widespread in most turkey-growing

areas that yearly vaccination of all turkey flocks is a good insurance policy. The one exception to this general recommendation is the flock well isolated from all chicken flocks and located in a community where the disease does not exist. Since the vaccine used for immunizing a flock contains live virus, capable of spreading the disease, it cannot safely be introduced into a flock or a community where fowl pox is unknown.

The disease is probably carried to new areas by mosquitoes, birds, visitors, animals, used feed sacks, and the introduction of new stock. Turkey growers who do not vaccinate should keep a constant watch for the first appearance



FIG. 40.20. Fowl pox lesions in the mucous membrane of the crop of a turkey. Lesions covered the head, and extended from the mouth to the crop in this bird. (Hinshaw, Univ. of Calif.)

of lesions and should immediately obtain advice on the best plan of control.

**Age for vaccination.** The correct age to vaccinate turkeys will depend on the locality. In some areas, it has been found necessary to vaccinate them by the end of June, regardless of age, because the prevalence of mosquitoes at that season spreads pox rapidly to all susceptible birds. In some communities it has been learned by experience that vaccination can be postponed until late in the fall, and in such instances it often is not necessary to revaccinate the breeders. There are considerable data to indicate that healthy turkeys can be vaccinated at any age. Dunn and Sherwood (1933) have successfully vaccinated day-old turkeys. Many growers have vaccinated successfully at six or eight weeks of age; the majority, however, at ten to twelve weeks. Extreme

care must be taken when vaccinating very young poults, to prevent the vaccine from getting on parts of the body other than the area to be treated. Sometimes a careless operator, after spilling vaccine, holds the poult's head with his contaminated hand. The young, tender skin is so susceptible that a severe case of generalized pox may follow.

As it requires from four to eight weeks for the vaccination lesion (take) to disappear completely, turkeys should be vaccinated at least eight weeks before market time.

Vaccines of chicken pox origin will produce immunity for about six months. It is, therefore, advisable to revaccinate all birds which are to be kept for breeders, six or seven months following the first vaccination. In order to be sure that the birds are no longer immune it is a good plan to revaccinate a sample (100 birds) of the flock. If after a week, the majority of the sample shows takes, the remainder of the flock can be revaccinated. If only a small percentage are susceptible, revaccination of the rest of the flock should be postponed. In some areas it may not be necessary to revaccinate because it has been learned from experience that the disease is not a factor during the breeding season.

*The wing web is not recommended as a site for applying the vaccine, because of disastrous results that have followed the use of this site.* One flock of eight-week-old poults, under the observation of the writer, which was vaccinated in the web of the wing by the puncture method suffered nearly a 50 per cent loss from fowl pox. Lesions developed on the head parts, in the mouth, and even in the mucous membrane of the upper esophagus, crop, and lower esophagus. Figure 40.21 A and B shows the results of wing web vaccination in an adult bird. At least 10 per cent of the flock from which the bird came was affected in a similar manner. Such spread of the infection is worse during damp or foggy weather and apparently associated with the birds picking at the vaccinated area before immunity develops.

The skin of the upper thigh has certain advantages over other sites for routine vaccination (Fig. 40.22). These are: easy accessibility to the operator, absence of feathers, and inaccessibility to the vaccinated birds or their penmates. The last point is important from the standpoint of the spread of fowl pox by fighting before immunity has been established.

The same methods of applying the vaccine are used for chickens and turkeys, and the reader is referred to the general section on fowl pox for these procdures.

**Reasons for failure in vaccination.** The chief reasons for failures are listed below:

1. Faulty care of vaccine after it leaves the producer.
   a. Undue exposure to heat in shipment.

    b. Improper care by the dispenser or by the purchaser.
2. Use of old vaccine kept beyond the producer's expiration date.
3. Improper use of diluted vaccine.
    a. Undue exposure to heat or sunlight.
    b. Failure to make up fresh supplies at short intervals.
    c. Failure to keep the vaccine well mixed.
    d. Attempts to economize by greater dilution than recommended by
       the producer.
4. Inefficient inoculation.
    a. Failure to dip the instrument into the vaccine after each inocula-
       tion.
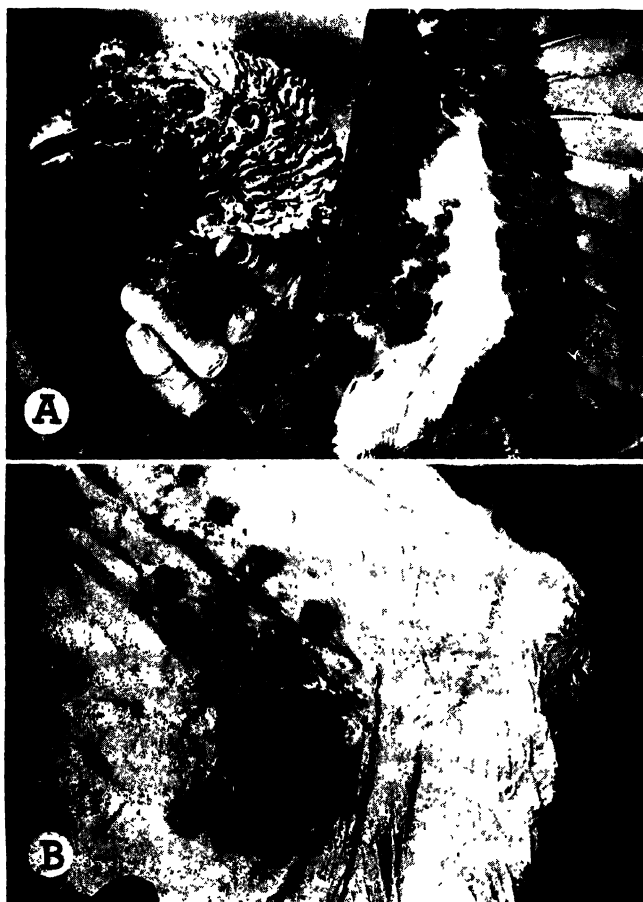


FIG. 40.21. A—wing web vaccination. Taken about six weeks after vaccinating in the wing web by a single stab of the inoculating knife. Shows how the disease spread from this single inoculation to other areas on the wing and head. The bird died within a few days after the picture was taken. B—close-up of A at a somewhat earlier stage. (Hinshaw, Univ. of Calif.)

b.  Failure to separate the feathers properly to expose the skin, with resultant loss of a large part of the vaccine as the instrument passes through the feathers.

c.  Failure to make an incision in the skin.

5.  Sacrifice of accuracy for speed.

6.  Previous immunity because of former outbreak or because of natural resistance.

**Control of an outbreak.** If fowl pox appears in a flock, the following procedure is recommended:

1.  Isolate all birds showing lesions.

2.  Vaccinate as soon as possible all birds not showing lesions.



FIG. 40.22. A—restraint of turkey for vaccinating on upper thigh. Note that the table is covered with newspapers. This aids in preventing undue spread of vaccine. B—a close-up, taken to show the suggested location. The long tuft of feathers that normally covers this naked area is being held back by the vaccinator's left hand. The heavy glass inkwell is a convenient holder for the vaccine. To prevent excessive dust contamination, it is covered with a piece of rubber with a small opening. (Hinshaw, Univ. of Calif.)

3. Place infected birds in warm, dry quarters if available.
4. Separate the males or keep careful watch over them to prevent fighting.
5. Treat infected birds individually by removing the scabs and touching the wounds with iodine, iodine ointment, or sulfonamide ointment, if time permits and the expense warrants it.
6. Individual feeding of valuable birds with the aid of a funnel and rubber tubing inserted into the crop may be advisable in severe cases.

**Drugs for internal treatment are not recommended.** Since loss of flesh and retarded development are the chief causes of economic loss in most outbreaks, careful management and attention to the feeding program during and after an outbreak are essential for a speedy return to normal. The course of the disease can be shortened by the use of shelter for roosting and by general protection from damp weather.

### REFERENCES

Beaudette, F. R., and Hudson, C. B.: 1941. Egg propagation of turkey pox virus. Poultry Sci. 20:79.

Blanc, G., and Caminopetros, J.: 1930. La transmission des varioles aviaires par les moustiques. Acad. Sci. (Paris) Compt. Rend. 190:954.

Brandly, C. A., and Dunlap, G. L.: 1938. An outbreak of pox in turkeys, with notes on diagnosis and immunization. Poultry Sci. 17:511.

Brody, A. L.: 1936. The transmission of fowl-pox. Cornell Univ. Agr. Exper. Sta., Memoir 195:1.

Brunett, E. L.: 1933. Some observations on pox virus obtained from a turkey. N. Y. St. Vet. Coll. Ann. Rep. 1932-33:69.

Coronel, A. B.: 1934. Fowl-pox vaccine from virus of turkey origin. Philippine Jour. An. Ind. 1:85.

Dunn, R. C., and Sherwood, R. M.: 1933. Immunization of day-old chicks and poults against fowl pox. Poultry Sci. 12:323.

Matheson, R., Brunett. E. L., and Brody, A. L.: 1931. The transmission of fowl-pox by mosquitoes (preliminary report). Poultry Sci. 10:211.

Tietz, G.: 1933. Ueber die Empfänglichkeit verschiedener Vogelarten für eine Infektion mit originärem Hühner-und Taubenpockenvirus. Arch. f. Tierheilk. 65:244.

### INFECTIOUS SINUSITIS
(Swellhead, Sinusitis)

This disease, which appears to be specific for turkeys, is characterized by inflammation of the lining membranes of the infraorbital sinuses, following which the sinuses become distended with a semigelatinous exudate (Fig. 40.23 A and B). Madsen (1938) and Hart (1940) have shown that the disease is transmissible by injection of exudate from infected into normal sinuses of turkeys. Neither was able to reproduce the disease by contact, and Hart failed also to reproduce it by either ocular or intranasal routes. Hinshaw and Bonestell (1940) have confirmed the results of these investigators. We could not produce the disease in chickens, but were successful in transmitting the disease back to turkeys by injection of saline washings removed from the sinuses of chickens given intrasinal injections a week previous:

Delaplane (1944) isolated a Pasteurella-like bacterium from exudate obtained from field cases of the disease in Texas. This organism produced

characteristic symptoms in chickens, and exudate from the sinuses of the chickens, when injected into turkeys, produced sinusitis. This fact would indicate that the disease he was dealing with has a different etiology than that studied by Hinshaw and Bonestell.

Hart states, "The causal agent . . . appears most likely to be a filtrable virus although two attempts to pass it through an Elford collodion membrane were unsuccessful." We were likewise unsuccessful in transmitting the disease with filtrates of sinus exudates passed through Chamberlain (L-5)
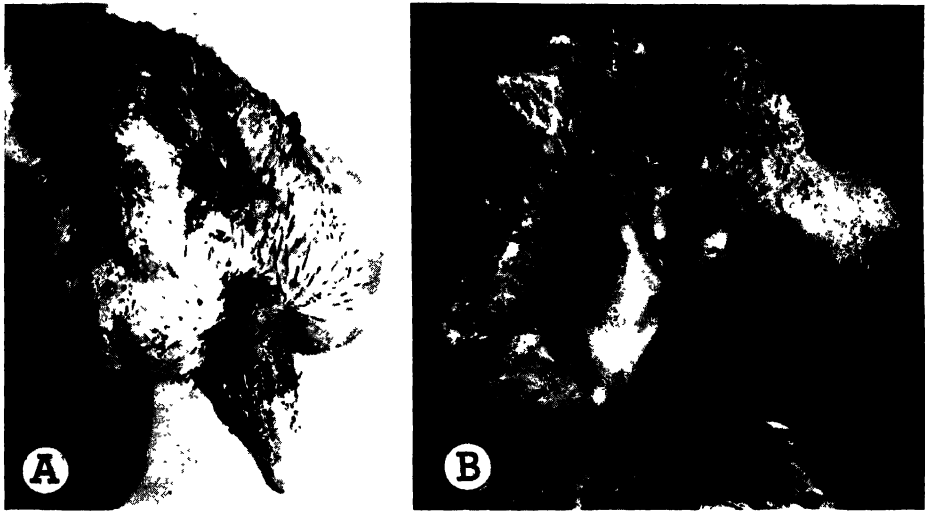


FIG. 40.23. A—an advanced case of infectious sinusitis involving both sides of the face. B—a similar case after the exudate in one sinus has been removed. (Hinshaw, Univ. of Calif.)

and sintered glass filters. A pleomorphic Gram-negative rod has been consistently isolated by us in pure culture. In only one instance, however, were we able to transmit the disease by injection of this organism into the sinuses of turkeys. In this one instance the first generation grown on horse blood agar was used, and there is the possibility that a filtrable factor may have been carried over on the medium.

The disease must be differentiated from sinusitis associated with vitamin A deficiency and from fowl coryza which Beach and Schalm (1936) found to be transmissible to turkeys. Mechanical injury caused by a piece of grain or mash or other foreign body becoming lodged in the sinus may result in a swollen sinus. As a rule these mechanical cases are unilateral. There is little question but that a diet deficient in vitamin A will predispose turkeys to this disease.

**Symptoms.** Prodromes of the disease are given when birds shake their

heads and when discharges are found on the feathers over the wing where the bird has attempted to clean its nostrils. These manifestations are followed by foaming of the eye secretions and by a clear nasal discharge. Swelling of the sinuses and, in advanced cases, a partial to complete closing of the eyes are the principal symptoms that follow these early signs. The appetite remains good as long as the bird can see to eat. As the disease progresses, the affected birds become thin but seldom show other symptoms. Labored breathing, in some cases, results from respiratory involvement or from complete closing of the palatine opening because of pressure from the exudate in the sinuses.

As these symptoms are characteristic for all types of sinusitis, final diagnosis depends on the history of the case and on autopsy findings.

**Course and mortality.** Sinusitis of the obviously contagious type runs a chronic course and may exist in a flock for weeks. Although the number of deaths may be less than in some more acute diseases, the financial loss may be greater. Failure to gain weight accounts for as much damage as does mortality.

**Autopsy findings.** The filling of the sinuses with exudate, the presence of pneumonia, and pleuritis are manifestations of this disease. Sinusitis usually occurs without involvement of the other respiratory passages, but in some flocks inflammatory changes in all the respiratory organs may be noted. In some instances, the lesions will be confined to the lower respiratory passages without involvement of the sinuses. Caseated exudate in the air sacs is common in acute outbreaks. When the lungs are affected, the bronchi are chiefly concerned.

The exudate in the sinuses in the first stages is watery in consistency, later becoming semigelatinous and finally caseated and whitish yellow in color. In typical outbreaks caseation of the exudate is the exception. In sinusitis associated with vitamin A deficiency, the lesions described under avitaminosis A will also be seen.

**Prevention, control, and treatment.** Since the exact cause of the disease is not known, no definite preventive recommendations can be given. As environment seems to play a part in its spread, protection of the flock from unnecessary exposure to drafts, windstorms, and sandstorms should be avoided. A diet adequate in vitamin A is also necessary.

Madsen (1938) has reported good results in the control of sinusitis by the use of 1.0 cc. of a 4 per cent solution of silver nitrate injected into the affected sinus after removal of the exudate with the aid of a hypodermic syringe. Dickinson and Hinshaw (1938), as well as Hart (1940), have used a 15 per cent argyrol solution in a similar manner. McNeil and Hinshaw (1946a) compared the efficiency of silver nitrate with three colloidal silver preparations, and two ephedrine remedies containing sulfonamides. The

latter were of no value, while the colloidal silver drugs were from 50 to 70 per cent effective as compared with about 85 per cent effectiveness of the silver nitrate.

The method consists in withdrawing the gelatinous exudate from the sinus with the aid of a 5 or 10 cc. syringe fitted with a 15- or 16-gauge needle, 1½ inches long. In the early stages of the disease this exudate is easily reached and removed when one learns the technic. It consists in inserting the needle through the skin and sinus membranes into the filled sinus. Withdrawal of the syringe plunger will remove the semifluid exudate. The needle is left inserted in the sinus, and with a second syringe the remedy (silver
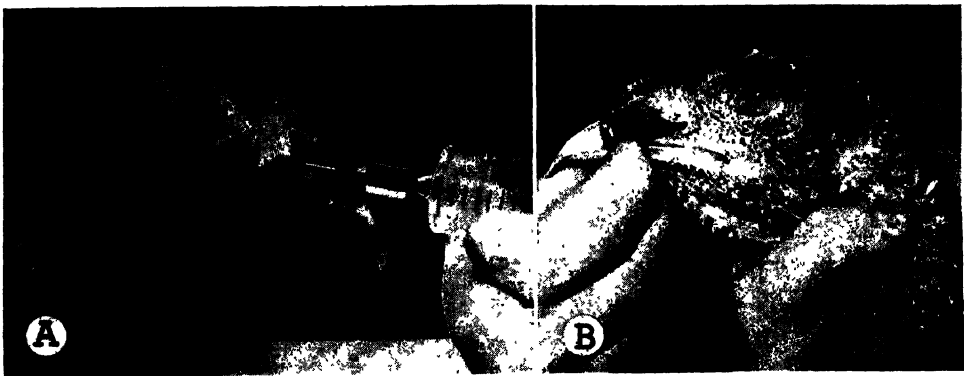


FIG. 40.24. A—treatment of infectious sinusitis. Method of insertion of the hypodermic needle into the sinus for withdrawal of exudate and inoculation of remedy. B—treatment of infectious sinusitis. After withdrawal of the exudate, the needle is left in the sinus until the therapeutic agent is introduced. (Hinshaw, Univ. of Calif.)

nitrate or argyrol) is injected and worked through the tissues by gentle massage. Care should be taken to avoid excessive dosage.

Both these treatments cause considerable swelling of the affected areas, but this subsides within 2 or 3 days, and complete recovery usually takes place within 10 days. In severe cases a second treatment may be necessary.

It is essential to administer this treatment in the early stages of the disease when the exudate is in a semigelatinous state. Figure 40.24 shows the method of inserting the needle for removal of the exudate, and for injection of the remedy.

No satisfactory treatment has been found for the birds that have an involvement of the respiratory organs (bronchi, lungs, and air sacs). Hinshaw and McNeil (1946) found that sulfathiazole, in addition to being unpalatable for young turkeys, was ineffective. Furthermore, this drug has a deleterious effect on egg production when given to laying flocks. Sulfamerazine was of some value if given at the rate of 0.5 per cent in the mash for 2 days and at the rate of 0.25 per cent for an additional 4 days (McNeil and

Hinshaw, 1946b) . This drug was effective only when given at the outset of an outbreak, and it did not prevent a recurrence of the disease.

If silver nitrate solution is used, it is advisable for the operator to wear leather or rubber gloves because this remedy is very caustic to the skin.

Surgical removal of the exudate and irrigation of the sinuses with a fresh solution of 15 per cent argyrol or 4 per cent silver nitrate solution may be necessary if the exudate has become caseous. A circular section of skin, at least 1/4 inch in diameter, over the swollen area should be removed; the exudate should be forced out by pressure with the thumb and forefinger. Following this, a piece of cotton saturated with the drug to be used can be inserted into the sinus to permit drainage and to prevent excessive dust collection. Care should be taken to avoid undue injury to the lining of the sinus. The treatment should be repeated every few days until improvement is noted.

It is a good plan, at the beginning of an outbreak, to send a representative specimen to a diagnostic laboratory in order to check on the possible presence of other diseases. There is no experimental evidence to substantiate claims that so-called "roup bacterins" will prevent or cure this disease. Until a specific cause is found and experimental evidence is available, bacterins cannot be recommended.

Although it is not definitely known that turkeys which recover from sinusitis remain carriers the following year, all contact between them and growing poults should be avoided. If a recovered flock must be used for breeding, the eggs should be hatched in an incubator, and the poults brooded artificially. As soon as enough eggs are obtained for producing the desired number of poults, the breeding flock should be brought into condition and sold for meat purposes to avoid possible contact with the poults during the rearing season. (Since this section was prepared, three groups of investigators have reported on the isolation of a filtrable virus from infectious sinusitis cases.)

## REFERENCES

Beach, J. R., and Schalm, O. W.: 1936. Studies of the clinical manifestations and transmissibility of infectious coryza of chickens. Poultry Sci. 15:466.

Delaplane, J. P.: 1944. A Pasteurella or Pasteurella-like organism as the cause of an infectious sinusitis of turkeys. Poultry Sci. 23:247.

Dickinson, E. M., and Hinshaw, W. R.: 1938. Treatment of infectious sinusitis of turkeys with argyrol and silver nitrate. Jour. Am. Vet. Med. Assn. 93:151.

Hart, L.: 1940. Sinusitis in turkeys. Australian Vet. Jour. 16:163.

Hinshaw, W. R., and Bonestell, A.: 1940.. Unpublished data.

—— and McNeil, E.: 1946. Experiments in the use of sulfathiazole for turkeys. Cornell Vet. 36:66.

Madsen, D. E.: 1938. Sinusitis of turkeys and its treatment. Utah Agr. Exper. Sta., Bul. 280.

McNeil, E., and Hinshaw, W. R.: 1946a. Recent studies on the treatment of infectious sinusitis in turkeys. Jour. Am. Vet. Med. Assn. 108:260.

—— and Hinshaw, W. R.: 1946b. Field trials on the use of sulfamerazine for a respiratory disease of turkeys. Poultry Sci. 25:273.

## FOWL TYPHOID

Fowl typhoid is a septicemic infection caused by *Salmonella gallinarum*. Cultures of this organism isolated from outbreaks among turkeys have resembled bacteriologically those isolated from chickens. Flocks grown in confinement have appeared more susceptible than those raised on open range; contact with chickens, or yards used by chickens, apparently has been an important factor in the spread of fowl typhoid to turkeys.

Pfeiler and Roepke (1917), Kaupp and Dearstyne (1924), Martinaglia (1929), and Hinshaw (1930) reported the disease in turkeys reared on farms where it was also prevalent in chickens. Evidence that the disease may be transmitted through the egg in the same manner as is pullorum disease is presented by Boney (1947), Hinshaw and Taylor (1933), and by Johnson and Pollard (1940). Johnson and Pollard reported outbreaks in poults with symptoms, mortality, and pathology comparable to those of pullorum disease. Usually, however, the disease is reported in mature or nearly mature turkeys. Vidovic (1931) claimed that strains of the causative organism isolated from turkeys were more pathogenic than strains isolated from chickens. The strains isolated from turkeys by Hinshaw have appeared identical to those isolated from other fowl. A more complete list of references is included in the general section on fowl typhoid.

**Symptoms, course, and mortality.** Increased thirst, loss of appetite, list-lessness, tendency to separate themselves from the well birds, and greenish to greenish-yellow diarrhea characterize the disease in the field. The sick turkeys sit with drooping tails, sagging wings, and heads hung low or carried back over the body and resting on or under the wing. As indicated by the increased thirst, the body temperature rises several degrees, to as high as 112° F., until just before death, when it may drop as low as 103° F.

Often birds die without having shown any previous symptoms, but usually they linger for a day or two after symptoms appear. Several outbreaks may occur in a flock in a single season, or the original one may be acute and last for only a few days. Intermittent outbreaks are more liable to occur if the birds are left on the originally infected premises or have constant contact with carrier chickens or turkeys. The initial outbreak usually causes the heaviest mortality, which is followed by intermittent recurrence of symptoms in a few birds, with a low mortality at each subsequent flare-up of the disease. Although the average mortality in four outbreaks studied was 26.5 per cent, heavier losses have often been reported. One flock owner lost 169 out of 175 turkeys during the fall and winter in intermittent outbreaks. In very young poults the symptoms, course, and mortality are similar to pullorum disease.

**Autopsy findings.** The lesions resemble those observed in chickens. Because of the short duration of the disease, the birds nearly always die while

in good flesh. The muscles of the breast have a tendency to be congested and often appear as if partially cooked. The heart is usually swollen and contains small, grayish necrotic areas or petechiae; in a few cases both have been observed. The liver is friable and is consistently enlarged to two or three times its normal size; it is bronze- to mahogany-colored or covered with a mixture of bronze- and mahogany-colored streaks. Pin-point areas of necrosis have been noted, though not consistently; on cutting the organ, the blood flows readily. The spleen is always enlarged to two or three times its normal size, is friable, and appears mottled. In most birds the lungs present a par-boiled appearance; often are more firm than normal, because of minute caseated abscesses. The kidneys are usually enlarged and may show some petechiae.

The crop, as a rule, contains food, which suggests paralysis of the digestive tract, since birds seldom eat after symptoms appear. The mucous membrane of the proventriculus sloughs readily. The gizzard usually contains food, and the lining is easily removed. With a few exceptions the intestine appears anemic when viewed from the exterior, and ulcerations of the mucous membrane may be plainly visible through the serosa. This ulceration is uncommon but when present is most severe in the duodenum; a few ulcers from 1.0 to 4.0 mm. in diameter have been observed throughout the intestine, extending to the ceca.

The enlarged mahogany- or bronze-streaked liver, the enlarged spleen, the area of necrosis in the heart, and the grayish lungs appear to be pathognomonic. Hemorrhagic enteritis, especially of the duodenum, and marked ulceration of the intestine, although uncommon in chickens, are more or less consistent lesions in turkeys. *Salmonella gallinarum,* the causative organism, can readily be isolated from all organs. In birds that have been dead for some time, pure cultures are more easily isolated from the bone marrow than from the liver, spleen, and heart blood.

In young poults Johnson and Pollard (1940) described the following autopsy findings: an increased percentage of large retained yolks, slightly enlarged somewhat friable liver of a white creamy color, with the surface mottled with slight hemorrhagic areas and slight congestion in the anterior duodenum. The crops, gizzards, and intestines were always devoid of food, indicating lack of appetite for several hours before death. In adult carriers, there is, as in the case of pullorum disease, a predilection for the reproductive organs (Fig. 40.25).

**Prevention, control, and treatment.** Since chickens are apparently the most common carriers of the disease to turkeys, the two species should never be allowed to mingle. It is equally important to keep turkeys from yards or ranges that have recently been used for chickens.

*Survivors of an outbreak should not be kept for breeders,* because of the

danger of transmission of the disease through the egg. If it is absolutely necessary to keep such survivors, the rigid program of multiple testing which is recommended for eradication of pullorum disease should be followed. The same test will detect carriers of both diseases.

Control depends upon eliminating the infection in the flock. The removal of all sick birds and the transfer of the well birds to a new range that has not been used for either chickens or turkeys is recommended. One
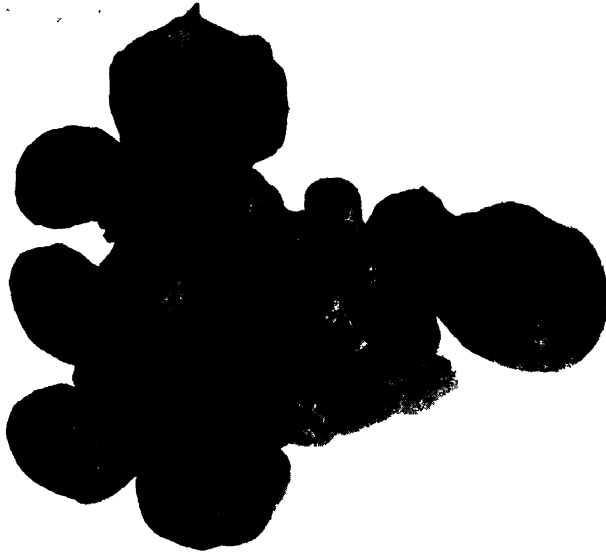


FIG. 40.25. Ovary from turkey hen, showing affected ova caused by fowl typhoid. No normal ova were present. (Hinshaw, Univ. of Calif.)

method for separating sick birds from well ones in an acute outbreak is to take the temperatures of all birds in the flock and eliminate those showing temperatures above 108° F. Another means of eliminating carriers, which has proved practicable in stopping intermittent losses, is to bleed the survivors and have the blood tested by means of the agglutination test. A diagnostic laboratory should be consulted before testing the flock.

Just before being moved, the birds should be given a laxative. A good laxative is a mash containing 40 per cent of dried skimmilk fed as the morning feed for 2 days. Epsom salt, if used, should not be given in doses exceeding 1 pound per 1,000 pounds of turkeys. Bran containing 5 per cent of molasses fed as a daily morning feed for a few days is another suggested laxative. Recent studies by Moore (1946), and by Holtman and Fisher (1946) indicate that certain of the sulfonamides are of value for reducing losses from fowl typhoid in chickens, if given before the disease becomes widespread. No reports are available on their value for turkeys, but they are worthy of a tiial.

(See discussion on the use of these drugs for turkeys under salmonellosis.)

As the greatest source of the spread of the disease is the droppings, the roosts should be screened to prevent the birds from having access to them. Sick birds should be taken out of the flock as soon as noted; frequent changes of the watering and feeding areas should be made; and whenever many new cases appear, the flock should be moved again to new quarters.

Next to the droppings, the greatest sources of infection are the food and water containers. They should, therefore, be cleaned and disinfected daily or even oftener. In the absence of running water, fresh, clean water should be given several times daily. Any antiseptic used should be one that will not make the water distasteful. Antiseptics that prevent the birds from drinking a normal amount of water do more harm than good.

Thus far experimental work has not demonstrated that vaccination with fowl-typhoid bacterins is effective for preventing or controlling the disease in turkeys.

## REFERENCES

Boney, W. A.: 1947. Isolation of *Shigella gallinarum* from turkey eggs. Am. Jour. Vet. Res. 8:133.

Hinshaw, W. R.: 1930. Fowl typhoid in turkeys. Vet. Med. 25:514.

—— and Taylor, T. J.: 1933. A chronic carrier of fowl typhoid of turkeys. Jour. Am. Vet. Med. Assn. 82:922.

Holtman, D. F., and Fisher, G.: 1946. Sulfa drugs in the control of *Shigella gallinarum* infections. Jour. Bact. 51:599.

Johnson, E. P., and Pollard, M.: 1940. Fowl typhoid in turkey poults. Jour. Am. Vet. Med. Assn. 96:243.

Kaupp, B. F., and Dearstyne, R. S.: 1924. Fowl typhoid—a comparison of various European strains with those of North America. Poultry Sci. 3:119.

Martinaglia, G.: 1929. A note on *Salmonella gallinarum* infection of ten-day-old chicks and adult turkeys. Jour. South African Vet. Med. Assn. 1:35.

Moore, E. N.: `1946. The efficacy of recently developed sulfonamides against fowl typhoid. Poultry Sci. 25:307.

Pfeiler, W., and Roepke, W.: 1917. Zweite Mitteilung über das Auftreten des Hühnertyphus und die Eigenschaften seines Erregers. Zentralbl. f. Bakt. I., Orig. 79:125. (Abst. in Jour. Comp. Path. and Therap. 30:263.)

Vidovic, F.: 1931. Die Untersuchungen der Pathogenität des *Bacterium gallinarum* bei Hühnern und Puten. Inaug-Diss. Vet. Fakult. Zagreb. 1930. Zentralbl. f. Bakt. Referate 103:472.

## NEWCASTLE DISEASE
### (Avian Pneumoencephalitis)

The reader is referred to the more complete discussion of this disease on pages 489 to 511. Turkeys are susceptible to Newcastle disease, and the symptoms and pathology are similar to those seen in chickens. The disease has not been as severe in commercial turkey-growing areas of the United States as it has in the chicken-producing areas. Newcastle disease may, however, become established in turkey-producing sections, so every effort should be made to locate outbreaks and to prevent its spread. Turkeys should not be reared in close proximity to chicken-rearing communities

where the disease is prevalent. If it is necessary to so rear them, vaccination of the turkeys should be considered. Recent literature should be consulted for the best preventive measures to follow, since intensive researches are now in progress on the disease.

## PARATYPHOID INFECTIONS
### (Salmonellosis)

This group of diseases is one of the major causes of losses, especially in young turkeys. This section deals with the types other than S. *pullorum* and S. *gallinarum*. There are now over 150 antigenic types of Salmonella described in the literature, and over 50 of these have been isolated from avian species. A check list of those found in turkeys follows:[1]

| | | | |
|---|---|---|---|
| S. *amherstiana* | S. *derby* | S. *meleagridis* | S. *san diego* |
| S. *anatum* | S. *dublin* | S. *minnesota* | S. *saint paul* |
| S. *bareilly* | S. *enteritidis* | S. *montevideo* | S. *senftenberg* |
| S. *berta* | S. *eastbourne* | S. *new brunswick* | S. *simsbury* |
| S. *bredeney* | S. *gaminara* | S. *newington* | S. *tennessee* |
| S. *california* | S. *give* | S. *newport* | S. *thompson* |
| S. *cerro* | S. *illinois* | S. *onderstepoort* | S. *typhimurium* |
| S. *chester* | S. *jabiana* | S. *oranienburg* | S. *typhimurium* |
| S. *cholera-suis* | S. *kentucky* | S. *oregon* | var. *copenhagen* |
| var. *kunzendorf* | S. *lexington* | S. *panama* | S. *urbana* |
| S. *concord* | S. *litchfield* | S. *paratyphi B* | S. *wichita* |
| S. *cubana* | S. *madelia* | S. *pomona* | S. *worthington* |
| | S. *manhattan* | S. *rubislaw* | |

The large list of Salmonella types now known to be capable of producing disease in turkeys becomes more significant when one considers that the first report of losses in turkeys was made as recently as 1933 (Rettger, Plastridge, and Cameron, 1933). Edwards (1939) reported the identification of eleven types sent to him for identification from thirty-one outbreaks. When the previous edition of this section was published in 1943, thirty-one types had been reported, and the disease had become of major importance as a cause of mortality in turkey flocks. Reference to types given in the checklist and not included by Edwards (1939) are made by Edwards and Bruner (1938, 1940, 1943); Pomeroy and Fenstermacher (1939, 1941a, 1941b); Edwards, Bruner, and Hinshaw (1940); and Hinshaw, McNeil, and Taylor (1944). Recently identified types not yet reported in the literature are from unpublished records of the University of Kentucky and the University of California. In addition to these reports from the United States, Saxer (1932) reported an outbreak caused by S. *enteritidis* (Gaertner type) after feeding turkeys meat from a calf suffering from navel infection. Nakamura, Nose, and Negishi (1939) also reported an outbreak in Japan caused by S. *enteri-*

---

[1] Compiled with the cooperation of Drs. P. R. Edwards, D. W. Bruner, and Miss Alice B. Moran, University of Kentucky, and Dr. Ethel McNeil, University of California.

*tidis.* A monophasic type having the antigenic formula IV,V,XII:e,h-, which is as yet unnamed, has been described by Cherry, Barnes, and Edwards (1946) and is not included in the checklist. This type was first isolated from a hog and later from turkeys and man. One outbreak of gastroenteritis in man caused by this type was traced to the eating of turkey meat.

It will be seen from this list that the problem of salmonellosis in turkeys is a complicated one, especially from the standpoint of eradication with the aid of the agglutination test, or control by use of bacterins. Of nineteen types found in California turkeys, *S. typhimurium* has accounted for about 60 per cent of the outbreaks (Hinshaw, McNeil, and Taylor, 1944). In other parts of the United States this type also appears to be the most important cause of losses in turkeys. The fact that *S. typhimurium* is the most common type found in turkeys complicates the problem even more because this type is also common to many other hosts, including other domestic fowl, wild birds, rodents, some farm animals, and even man. Many of the types found in turkeys have been found first in food poisoning outbreaks in man. Therefore, any plan for the control and prevention of salmonellosis in turkeys must include the control in man and other animals as well.

**Transmission.** Evidence that this group of diseases may be transmitted through the egg in a manner similar to pullorum disease has been presented by Cherrington, Gildow, and Moore (1937), who succeeded in isolating *S. typhimurium* from two of six ovaries removed from reacting turkeys, and from three of thirty "dead-in-shell" embryos. Lee, Holm, and Murray (1936) in an earlier paper reported the isolation of this organism from four of ten ovaries removed from artificially infected turkey hens. At this station (California), *S. typhimurium* has been isolated from both the ovary and oviduct of turkey hens (Hinshaw and McNeil, 1943).

The incidence of *S. typhimurium* in eggs laid by carriers is not high according to the literature available and our own experience. Pomeroy and Fenstermacher (1939) report the isolation of paratyphoid organisms from 7 out of 200 incubated eggs that failed to hatch. *S. typhimurium* was isolated from the ovaries of two of the nine reactors that laid the above-mentioned eggs. In a later paper (1941a) these investigators showed that *S. typhimurium* will pass through the unbroken egg shell, and infect developing embryos, some of which hatch and become a source of infection to normal poults. Schalm (1937) earlier showed that this same method of infection is possible in chicken eggs. Therefore, in addition to ovarian transmission, transmission by eggs contaminated by fecal matter during expulsion from the body, and in the nest, is possible. From information available, it would seem that this method is more common than by the former method. Hinshaw and McNeil (1943) reported that 81 per cent of adult *S. typhimurium* car-

riers yielded the organism from the intestines, in contrast to 17 per cent which yielded it from the reproductive organs.

**Symptoms.** The symptoms in young poults are indistinguishable from pullorum disease. The age at which poults may be affected ranges from a few days after hatching to maturity. In general, however, the age incidence is that of pullorum disease—from 3 or 4 days of age to one month. The age at which the disease is first observed in poults will depend on whether the poults are infected while in the incubator or after being placed in the brooder. In one outbreak studied, symptoms were seen 2 days after the poults were taken from the incubator, indicating transmission in the incubator soon after the eggs start to hatch.

Diarrhea in young poults is not a constant symptom; often poults normal in the evening may be found dead in the morning. Where death is delayed for several days, weakness, unthriftiness, sagging wings, and diarrhea are characteristic symptoms. Many poults that survive for several days will become emaciated, and the feathers around the vent will be matted with fecal material.

In older turkeys, loss of appetite, unthriftiness, loss of flesh, and a general unkempt appearance have been the symptoms most commonly observed. Diarrhea may or may not be in evidence. Death usually follows after several days of sickness.

**Autopsy findings.** Inflammation of the duodenum, congestion of the liver, kidney, gall bladder, and heart muscle are the most constant post-mortem findings. The pericardial sac is often filled with a serous straw-colored fluid. Another common finding is a cecal plug similar to that sometimes seen in pullorum disease. Lung and heart lesions are rare, but air sac involvement is common.

In adult turkeys, marked inflammation of the intestine with occasional necrotic ulcers is seen. The liver and spleen in these cases is usually swollen and congested. Diagnosis depends on isolating and identifying the causal organism.

**Prevention and control.** Prevention consists in obtaining stock which is free of the disease and in preventing the birds from being exposed to other animal reservoirs of infection. As stated before, most of the types of Salmonella repsonsible for losses in turkeys are also prevalent in other animals and even in man. Thus the program of prevention must be extended to all animals on the ranch. Eradication of rats, mice, flies, and reptiles is essential in the prevention of this group of diseases. If the disease is diagnosed in any group of poults, the poults should be reared separately from other groups, and sold for market. Such infected groups should never be used for breeders.

The use of the agglutination test as an aid in determining the presence or

absence of the disease on the premises, will depend on the facilities available for having such tests made. If a diagnosis of the disease has been made and the type of Salmonella determined by a reliable laboratory, the agglutination test may be used if made by a laboratory thoroughly familiar with the problems connected with conducting paratyphoid agglutination tests. Each ranch must be handled as an individual unit when making plans for testing. A separate test must be made for each species isolated, and complete knowledge of the disease history of the flock is essential to a successful program. The agglutination test for the paratyphoids is more complicated and, as now conducted, more subject to variation than is the one for pullorum disease. To conduct it properly, the laboratorian must be thoroughly familiar with the antigenic structure of Salmonella, and be able accurately to interpret results. On ranches where the infection is known to exist, the test may be a valuable adjunct in the prevention program, but must be considered only as one part of such a program. It is absolutely necessary to know the type causing the disease, and as may often be the case, there may be two or more species responsible (Edwards and Bruner, 1940). If the complete history is known and a competent laboratory is available, testing is advised (Hinshaw and McNeil, 1943). A general testing program for the paratyphoids will probably never be possible because of the multiplicity of types affecting turkeys. Every effort should be made to prevent any of the paratyphoids from getting into breeding flocks. The destruction of broods suffering from the disease, and the purchase of replacements from sources known to be free is one plan which can be used.

Hatcheries and egg-selling groups can help prevent the group of diseases from spreading by keeping thoroughly familiar with all the ranches furnishing hatching eggs. Frequent use of the diagnostic laboratory is urged during the brooding season in order to insure a high percentage of diagnoses of the outbreaks of paratyphoid which occur. Whenever a diagnosis is made the owner should be made familiar with the problem and responsibility he has in preventing the spread of the disease. Often, only one brood is affected, and it will be good insurance to destroy all the survivors. In any case survivors of an outbreak normally should not be used for breeding purposes. If the flock has to be used for breeders, it should be tested, using the same type isolated, and retested at frequent intervals until no reactors are found. Competent advice should be sought to determine the proper procedure to follow. *Other animal and bird reservoirs on the ranch must also be eliminated if the disease is to be eradicated.* Cats, flies, and even snakes and lizards are important carriers of this group of diseases, and should not be overlooked in outlining the control program (McNeil and Hinshaw, 1944; Hinshaw and McNeil, 1945; Hinshaw and McNeil, 1947).

Recent reports by Clark (1946), and Pomeroy, Fenstermacher, and

Roepke (1946) indicate that sulfamerazine, sulfadiazine, and sulfamethazine have value for reducing mortality, when given at the rate of 0.25 to 0.5 per cent of the mash for periods not exceeding one week. In general the sulfonamides are more toxic for poults than for chicks, and must, therefore, be used with caution. These drugs have not proven of value in eliminating carriers from an infected flock. West, Doll, and Edwards (1946) studied the effect of streptomycin in vitro on 412 cultures of all the recognized types of Salmonella. The majority of the strains required from two to four times as much streptomycin to inhibit growth as did a standard test strain of *E. coli*. Until more is known about the efficiency of bacterins for prevention and control, they cannot be recommended. The multiplicity of types makes the general use of bacterins as impossible as a generalized testing program at the present time.

## REFERENCES

Cherrington, V. A., Gildow, E. M., and Moore, P.: 1937. Paratyphoid in turkeys. Poultry Sci. 16:226.

Cherry, W. B., Barnes, L. A., and Edwards, P. R.: 1946. Observations on strains of a monophasic Salmonella variant. Jour. Bact. 51:235.

Clark, C. H.: 1946. Sulfamerazine in paratyphoid disease of poults and chicks. Jour. Am. Vet. Med. Assn. 109:279.

Edwards, P. R.: 1939. Incidence of Salmonella types in fowls in the United States. Proc. Seventh World's Poultry Cong.: 271.

—— and Bruner, D. W.: 1938. Two new Salmonella types isolated from fowls. Jour. Hyg. 38:716.

—— and Bruner, D. W.: 1940. The occurrence of multiple types of paratyphoid bacilli in infections of fowls, with special reference to two new Salmonella species. Jour. Infect. Dis. 66:218.

—— and Bruner, D. W.: 1943. The occurrence and distribution of Salmonella types in the United States. Jour. Infect. Dis. 72:58.

——, Bruner, D. W., and Hinshaw, W. R.: 1940. A new Salmonella type isolated from turkeys: *Salmonella california*. Jour. Infect. Dis. 66:127.

Hinshaw, W. R., and McNeil, E.: 1943. The use of the agglutination test in detecting *Salmonella typhimurium* carriers in turkey flocks. Proc. Forty-Seventh Ann. Meet. U. S. Livestock Sanitary Assn.: 106.

—— and McNeil, E.: 1945. Salmonella types isolated from snakes. Am. Jour. Vet. Res. 6:264.

—— and McNeil, E.: 1947. Lizards as carriers of Salmonella and paracolon bacteria. Jour. Bact. 53:715.

——, McNeil, E., and Taylor, T. J.: 1944. Avian salmonellosis. Types of Salmonella isolated and their relation to public health. Am. Jour. Hyg. 40:264.

Lee, C. D., Holm, G., and Murray, C.: 1936. Paratyphoid infection in turkeys. Jour. Am. Vet. Med. Assn. 89:65.

McNeil, E., and Hinshaw, W. R.: 1944. Snakes, cats, and flies as carriers of *Salmonella typhimurium*. Poultry Sci. 23:456.

Nakamura, N., Nose, Y., and Negishi, B.: 1939. An outbreak of *Salmonella enteritidis* infection in baby turkey poults. Proc. Seventh World's Poultry Cong.: 240.

Pomeroy, B. S., and Fenstermacher, R.: 1939. Paratyphoid infection of turkeys. Jour. Am. Vet. Med. Assn. 94:90.

—— and Fenstermacher, R.: 1941a. Paratyphoid infection of turkeys. Am. Jour. Vet. Res. 2:285.

—— and Fenstermacher, R.: 1941b. Salmonella infection of breeding turkeys. Abst. Jour. Am. Vet. Med. Assn. 99:216.

——, Fenstermacher, R., and Roepke, M. H.: 1946. Sulfonamides in pullorum disease of chicks and poults. Vet. Med. 41:438.

Rettger, L. F., Plastridge, W. N., and Cameron, R.: 1933. Endemic paratyphoid infection in turkeys. Jour. Infect. Dis. 53:272.
Saxer, E.: 1932. Gärtnerinfektion bei Truthühnern. Schweizer Archiv für Tierheilk. 74:351. (Abst. from Internat. Rev. Poultry Sci. 3/4:96. 1933.)
Schalm, O. W.: 1937. Study of a paratyphoid infection in chicks. Jour. Infect. Dis. 61:208.
West, M. G., Doll, E. R., and Edwards, P. R.: 1946. Inhibition of Salmonella cultures by streptomycin. Proc. Soc. Exper. Biol. and Med. 60:363.

## PULLORUM DISEASE
### (Bacillary White Diarrhea, White Diarrhea)

This disease, caused by *Salmonella pullorum*, has been increasing in economic importance to the turkey industry since the advent of the commercial hatching of turkey eggs. It was first described in turkeys by Hewitt (1928) in Minnesota and has since become widespread among turkeys in America as well as in some of the other countries of the world. Comprehensive reviews of the literature have been given by Tittsler (1932), Johnson and Anderson (1936), and Hinshaw (1939). Investigators in other countries who have reported on the disease in turkeys include Dalling, Mason, and Gordon (1929), Jansen (1932), and Barboni (1937).

Previous to 1938 when Johnson and Anderson reported an outbreak which apparently originated from eggs laid by turkey carriers, all the evidence pointed to chickens as the main source of the infection in turkeys. The disease is now, however, well established in many turkey flocks, and the cycle of infection has been shown to be identical to that for chickens (Hinshaw and McNeil, 1942).

**Symptoms.** The symptoms in poults are identical with those described for chicks. The disease is usually very acute, and many poults die without showing any noticeable symptoms. Poults that show symptoms seem cold and sit around the hot part of the hover space. Their wings sag, their heads hang, and their feathers appear unkempt. The skin over the feet and legs usually appears dry and somewhat wrinkled. Diarrhea may or may not be present; but in most of the cases that are prolonged for 2 or 3 days, diarrhea is indicated by the pasting of the down around the vent. Labored breathing, due to pneumonia, is commonly observed.

**Course and mortality.** Most of the losses occur during the first three weeks after hatching, and may start as early as the second day. It is not uncommon for relapses to occur at any time up to maturity, with varying degrees of mortality. Frequently, when survivors of an early age outbreak reach nine to ten weeks of age and are transferred to a growing ration or moved to new quarters, a second outbreak occurs with a subsequent mortality of 5 to 15 per cent. Such relapses may also occur during outbreaks of hexamitiasis. The diagnostic laboratories of the California State Department of Agriculture have reported several outbreaks in turkeys three to six months of age. Losses of this age group have, as a rule, been small. Subacute outbreaks have

also been experienced in breeding flocks after they are in production. Such outbreaks are attributed to transmission by eating infected eggs and subsequent spread by the intestinal shedders of the organisms.

The mortality in poults under one month of age varies from less than 10 to as high as 100 per cent of a brood, depending on the virulence of the organism, and the management of the brood.

**Autopsy findings.** There are a few distinctive changes in poults that have died from pullorum disease. Minute caseous abscesses in the lungs and heart muscles similar to those seen in chicks are the most characteristic lesions. Similar abscesses may be found in the gizzard muscles. The intestines usually lack tone, and contain an excessive mucous discharge. Cecal cores are seen, but they are not pathognomonic for this disease alone. The liver is commonly congested and swollen or may be of an ocher to a bronze color streaked with areas of congestion. Pin-point areas of necrosis are also common.

The post-mortem findings in partially grown poults are similar to those seen in younger ones, but these are usually less pronounced. Lung lesions are only occasionally seen, but necrotic foci in the liver are frequent findings, as are nodules in the gizzard and catarrhal enteritis.

The lesions seen in adult carriers are similar to those seen in carrier chickens, and are principally confined to the reproductive tract. *S. pullorum* has also been isolated occasionally from the lungs, intestines, bursa of Fabricius, and liver, and in one instance from the testes of reactors. A frequent finding in adults that have been killed in subacute outbreaks is marked ascites. In these cases as much as 1,000 cc. of fluid containing yellowish caseated flaky masses may be removed from a single bird. As a rule, such specimens yield *S. pullorum* from all tissues including the intestines.

**Prevention.** Now that pullorum disease has become widespread in turkey flocks, and turkey carriers are important means of transmission, both turkey and chicken carriers (as well as other fowl carriers) must be eliminated if the disease is to be prevented. The *first prerequisite* in a program of prevention is to establish a source of pullorum-disease-free eggs. The *second* is to have such eggs hatched in a hatchery that accepts eggs only from pullorum-disease-free flocks of turkeys, chickens, and other fowl. The *third* is to brood and rear the poults in brooders with equipment that has had no contact with birds of any species that have had pullorum disease. If these three prerequisites are followed together with a good management program, there is little danger that pullorum disease will become established in a flock of turkeys.

When an outbreak occurs, it is recommended that the survivors be marked and reared separately from broods that have not had the disease. Such groups of survivors should be sold for market, and never kept for breeders. They should be marketed before they start to lay eggs. The remainder of the birds on the premises, if they are to be kept for breeders, should be tested by

means of the tube agglutination test. According to Hinshaw and McNeil (1942) this test, using a 1:25 dilution, made according to the procedure recommended in the National Turkey Improvement Plan (1946) is a reliable aid in locating pullorum-disease-free flocks, and in eradicating the disease from infected flocks. Hinshaw, Jones, Harr, and Niemeyer (1940) reported that the whole-blood stained antigen test was 50 per cent as efficient as the standard tube test for detecting carriers of *S. pullorum*,. Bushnell (1945) , and Corpron, Bivins, and Stafseth (1947) have also reported that the tube test is more sensitive and consistent than the more recently developed whole-blood stained antigens. Gauger (1947) reported close agreement between the whole-blood K-antigen and the tube test when the titer of the latter was complete agglutination in 1:25 dilution or better.

The procedure to be recommended for eradication of the disease from infected ranches will depend on many factors, and every ranch must be considered an individual problem when mapping out a program. Factors which have to be considered are (1) type of manager, (2) plan of ranch, (3) type of available equipment, (4) size of yards, (5) drainage, and (6) history of flock. *In general, the best recommendation, if infection is known to exist, is to sell the entire flock and replace it with stock from sources known to be free of the disease.* If only one age group is known to be infected, the sale of this group for market may suffice, providing the remainder of the flock is tested and found to be without reactors. The average percentage of reactors found on the initial test of a flock is not in itself sufficient information to enable one to evaluate subsequent procedure.

Under no condition is any percentage of infection above zero (equivalent to the U. S. Pullorum Clean Class, or the U. S. Pullorum Passed Class) to be tolerated if pullorum disease is to be eradicated from turkey flocks. Furthermore, hatcheries that accept any eggs (chicken, turkey, or other birds) from sources which are not known to be free of the disease, cannot be tolerated if eradication is to be accomplished.

It is possible to completely eradicate infection from a flock in one year, but as stated above many factors must be considered. If an infected flock is to be used for production of hatching eggs, the following procedure should be carried out: (1) Make the initial test when the flock is four to five months of age; (2) divide the flock into small groups on clean ground two weeks before testing; (3) remove all reactors promptly and dispose of them immediately; (4) thoroughly clean and disinfect all feeding and watering equipment; (5) dispose of all infected pens or remove them to clean ground as soon as reactors are removed; (6) retest infected pens once a month until free of reactors if they are retained; (7) after all groups are free by the pen method, retest the entire flock in one month to ascertain if any new reactors have developed; (8) use eggs for the breeding flock replacements, only from

free-on-first-test sources; (9) have all eggs for replacements hatched by a hatchery accepting only eggs of like status; (10) practice a rigid sanitary program at all times. As stated above, the exact procedure will vary with the individual ranch situation.

It is often necessary to keep untested birds on the premises until they can



FIG. 40.26. A—type of catching chute for handling large numbers of turkeys for vaccination, blood collection, or other purposes. B—the same arrangement showing how a bird can be removed from the side, by reaching through the burlap "fence," without disturbing the other turkeys. (Hinshaw, Univ. of Calif.)

be marketed. If complete segregation can be maintained, this may be done. Again the individual ranch situation will influence the recommendations to be made.

*It cannot be emphasized too often that the hatchery is the keynote of any eradication program. A single noncooperating hatchery can destroy the results of an entire program by accepting eggs that are not of equal status with those cooperating in the program. Likewise, a noncooperating grower can be responsible for spread of infection, by failing to observe the above*

*recommendations.* This is especially true in cooperative groups where pooled eggs from several ranches are often used for furnishing replacements to a single grower. In such organizations, the quality of the cooperative group's eggs is the quality of those of its poorest cooperator.

**Bleeding of the flock for the agglutination test.** Since the breeding flock can usually be selected early in the fall and separated from the birds to be marketed, the turkey growers should do this before bleeding for the first test. At least 25 per cent more turkeys than are needed for the breeding flock should be separated to allow for losses caused by removal of the reactors to the test, and culling for breed improvement. Males as well as females should be tested; and any chickens or other fowls kept in the vicinity should also be tested.

Much the same arrangement as was suggested for handling turkeys for fowl pox vaccination may be used in corralling the flock (Fig. 40.26). One should always consult the laboratory which is to do the testing for special procedures recommended by it. The type of vial used, the amount of blood desired, the



FIG. 40.27. Pullorum disease. Method of withdrawing a blood sample from the median vein of the wing. (Hinshaw, Univ. of Calif.)

type of shipping container, and the method of shipping the samples to the laboratory are factors which may vary. If an official program is in operation, it may be necessary to make special arrangements, and it is always advisable to make application for a definite date several weeks before the testing is desired. The instructions of the laboratory which is to make the test should be carefully followed with reference to identification of the birds, collection of blood, its subsequent handling, and shipment to the laboratory.

Two general methods are used for collecting the blood samples. One of these is by use of a hypodermic syringe and needle according to the method described by Van Es and Olney (1941). The second is by the so-called stab or "nick" method in which the wing vein is punctured with the aid of a sharp-pointed knife such as a Bard-Parker number 11. The "syringe method" of bleeding turkeys is the recommended method. By this procedure, it is possible to collect more uniform samples which are cleaner, and which reach the laboratory in much better condition than those collected by the "nick" technic. The technic for bleeding turkeys by the syringe. method, is essen-

FIG. 40.28. Pullorum disease. Equipment used by a bleeding crew for collecting blood samples by the syringe technic. (Hinshaw, Univ. of Calif.)

tially that described by Van Es and Olney. Luer-lok type 2 cc. syringes and 18 gauge needles are preferred. Approximately 1 cc. of blood is ample and can be collected by inserting the needle into the median vein of the wing (*Vena cutanea ulnaris*) as illustrated in Figure 40.27. To work efficiently, it is desirable to have several sets of syringes and needles, so that an assistant can keep a supply ready for the operator at all times. After the sample is

drawn, an assistant records the number of the bird, empties the blood into the vial, rinses the syringes at least three times, and lays it down in a convenient place for the operator. Three rinse waters are used; the first, tap water; the second and third, physiological saline. These rinse waters should be boiled before use and should be changed frequently. If the syringes are well rinsed and these precautions followed, the method is superior to the "nick" technic (Figs. 40.28 and 40.29).

An efficient crew consists of the operator, and at least two assistants: one for banding the birds, and one
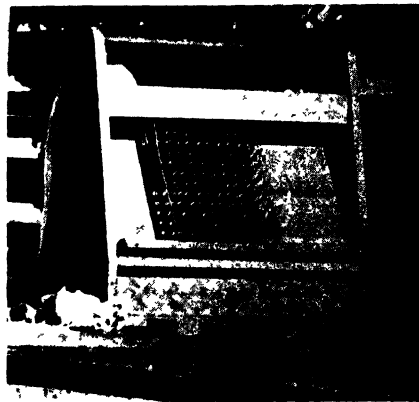


FIG. 40.29. Type of rack used by the California Poultry Improvement Advisory Board for holding blood samples during the bleeding procedure. (Hinshaw, Univ. of Calif.)

for recording the band numbers, emptying the syringes, and cleaning them. If speed is to be maintained, the operator should only have to withdraw the blood. It is, therefore, necessary that the flockowner furnish ample help in order always to have a bird on the table ready to be bled. Figure 40.30 illustrates a few of the steps in this technic.

**Control and treatment.** There is no practical method of control or treatment once the disease has become established in a brood. Daily cleaning and the removal of all sick and dead poults several times daily will aid in



FIG. 40.30. Pullorum disease. A crew at work bleeding a flock of turkeys. (Hinshaw, Univ. of Calif.)

preventing its spread. Increasing the heat in the brooder may be helpful in preventing excessive loss. Cleaning and disinfecting the water fountains and feed hoppers several times daily and the use of fresh, unadulterated water are also recommended.

Recently, sulfonamides have been given much publicity for control of pullorum disease outbreaks in chicks and poults. Severens, Roberts, and Card (1945), Bottorff and Kiser (1947), Mullen (1946), and Anderson (1946) have shown that mortality can be reduced by the use of sulfonamides given at the rate of 0.25 to 0.5 per cent in the mash for periods up to a week. Drugs tried by these investigators include sulfamerazine, sulfadiazine, and sulfamethazine. Complete prevention of losses have not been reported. Contrary to the early impressions created by results obtained by Roberts, Card,

and Severens, carriers are not eliminated by sulfa drug treatment, so their use is not a substitute for a testing program. Treatment with sulfonamides should be made only after a positive diagnosis has been made, and then only upon the advice of a competent veterinarian. Infected flocks that have been saved by treatment should not be used for breeding purposes.

Every precaution should be taken to prevent contact of an infected brood with other broods that are to be brought into the house after the outbreak is in progress. The brood suffering from the disease should be kept in isolated quarters. Under no circumstances should equipment used for the infected brood be used for later hatches until it has been thoroughly cleaned and disinfected.

When the disease has run its course, the survivors should be toe-marked and raised separately from the other lots. *None of the survivors should be saved for breeding purposes.* The survivors should be marketed as soon as they are in condition and the breeders selected from groups that have not suffered from the disease. These breeders should be tested as described under prevention.

## REFERENCES

Anderson, G. W.: 1946. Sulfamerazine in the treatment of pullorum disease. Jour. Am. Vet. Med. Assn. 108:427.

Barboni, E.: 1937. Ricerche sul primo focolaio di pullorosi nei tacchini riscontrato in Italia. La Clin. Vet. 60:597.

Bottorff, C. A., and Kiser, J. S.: 1947. The use of sulfonamides in the control of pullorum disease. Poultry Sci. 26:335.

Bushnell, L. D.: 1945. Pullorum testing of turkeys. Poultry Sci. 24:208.

Corpron, R., Bivins, J. A., and Stafseth, H. J.: 1947. Pullorum disease studies in turkeys. Poultry Sci. 26:340.

Dalling, T., Mason, J. H., and Gordon, W. S.: 1929. Bacillary white diarrhea (B.W.D.): B. pullorum isolated from a turkey poult in England. Vet. Rec. 9:902.

Gauger, H. C.: 1947. Comparison of the rapid whole-blood K-antigen and the tube agglutination test for the detection of pullorum disease in turkeys. Poultry Sci. 26:229.

Hewitt, E. A.: 1928. Bacillary white diarrhea in baby turkeys. Cornell Vet. 18:272.

Hinshaw, W. R.: 1939. Diseases of turkeys in United States—a review. Proc. Seventh World's Poultry Cong.: 236.

————, Jones, E. E., Harr, J. F., and Niemeyer, W. E.: 1940. Comparison of the tube and the whole blood tests for pullorum disease of turkeys. Cornell Vet. 30:30.

————, McNeil, E., and Taylor, T. J.: 1942. Four years progress in eradication of pullorum disease from turkey flocks. Proc. Forty-Sixth Ann. Meet. U. S. Livestock Sanitary Assn.:224.

Jansen, J.: 1932. Chronische pulloium-infectie bij volwassen kalkoenen. Tijdschr. voor Diergeneesk. 59:1047.

Johnson, E. P., and Anderson, G. W.: 1936. Pullorum disease in turkeys. Jour. Infect. Dis. 58:337.

Mullen, F. E.: 1946. Sulfamerazine as a prophylactic in pullorum disease. Jour. Am. Vet. Med. Assn. 108:163.

Severens, J. M., Roberts, E., and Card, L. E.: 1945. The effect of sulfonamides in reducing mortality from pullorum disease in domestic fowl. Poultry Sci. 24:155.

The National Turkey Improvement Plan: 1946. U.S.D.A. Misc. Publ. 555.

Tittsler, R. P.: 1932. Pullorum disease in poults. Poultry Sci. 11:78.

Van Es, L., and Olney, J. F.: 1941. Poultry diseases and parasites. Nebr. Agr. Exper. Sta., Bul. 332:42.

## SPIROCHAETOSIS[*]

Until Hoffman, Jackson, and Rucker (1946) reported an outbreak of spirochaetosis in turkeys in California, the disease was not known to exist in North America. No vector could be incriminated in the California outbreak although a careful search was made. Since then Burroughs (1947) has reported a case of spirochaetosis in a fowl which was used for feeding fowl ticks (*Argas persicus*) obtained from a poultry flock in Texas. Burroughs' findings would indicate that the disease may be prevalent in chickens in Texas even though it was not previously reported. Steinhaus and Hughes (1947) reported the isolation of a nonpathogenic spirochaete from hen eggs following inoculation with liver tissues from chickens. Their spirochaete is not *Borrelia anserina* (Sakharoff) (=*Spirochaeta gallinarum*, Blanchard), the causative agent of fowl spirochaetosis. In a later paper Hoffman and Jackson (1946) reported more completely on the same outbreak referred to above. Hinshaw and McNeil (1946) studied the spirochaete obtained from Hoffman's outbreak and found it to have all the characteristics of *Borrelia anserina*.

Approximately a year after the outbreak referred to by Hoffman *et al.*, occurred, another outbreak was diagnosed by the University of California, Veterinary Department Laboratories (unpublished), in a flock of adult turkeys located about 150 miles from the original outbreak. As far as is known this is the only new outbreak which has occurred. As was the case in the original outbreak, fowl ticks could not be found on the infected ranch. The spirochaetes obtaind from the second outbreak have been compared with those from the first outbreak and proved to be identical.

Fowl spirochaetosis is widely distributed over the world. For a more complete worldwide review the student is referred to van Heelsbergen (1929), Reis and Nobrega (1936), and Lesbouyries (1941). Recent reviews of the literature are given by Sreenivasan and Sankaranarayan (1945) and Morcos, Zaki, and Zaki (1946). The only published reference to a natural outbreak in turkeys, other than that of Hoffman *et al.*, is one by Stylianopoulos (1925) which is referred to by Lesbouyries (1941). This outbreak occurred in Greece in turkey poults.

**Vectors.** Although the fowl tick *Argas persicus* is generally referred to as the vector for *Borrelia anserina*, it is by no means the only vector. Others that have been reported include the common red mite *Dermanyssus gallinae* by Hungerford and Hart (1937), and Culex mosquitoes by Zuelzer (1936). Direct transmission is also possible by several routes including oral, intranasal, intraorbital, intravenous, and subcutaneous. Kapur (1940) was able

---

[*]A considerable portion of this section is based on research work to be published by Hinshaw, McNeil, and Kissling.

to transmit the disease in chickens by smearing the infected material on the unbroken skin of the comb or the breast. The incubation period in such cases was 2 to 6 days.

**Symptoms.** Listlessness, cyanosis of the head, increased thirst, fever, and yellowish-green diarrhea with increased urates are characteristic symptoms. The area around the vent is nearly always stained with urates. In turkeys artificially infected by the intravenous route the body temperature usually increases within 24 hours following infection, and reaches a peak of 109.0° F.



Fig. 40.31. Spirochaetosis. Typical attitudes seen in acute outbreaks in turkey flocks. (Univ. of Calif.)

to 111.0° F. on the fourth or fifth day. By the end of the seventh or eighth day, if the bird lives, the temperature usually returns to normal. In naturally developed cases temperatures as high as 109.4° F. have been observed. Infected birds tend to sit with their eyes closed unless disturbed. Chronically affected individuals develop leg weakness and sit characteristically on their hocks (Fig. 40.31). When disturbed they move about by hopping, often in a semisquatting position rather than on their feet. Others walk with a stilted gait. Complete paralysis has occasionally been noted.

**Autopsy findings.** The most characteristic gross change noted on autopsy is a marked enlargement and mottling of the spleen, due to ecchymotic hemorrhages such as are seen in Fig. 40.32. The heart may be enlarged and have a parboiled appearance. The liver is usually enlarged, congested, and more or less studded with minute areas of necrosis. In advanced cases the areas of necrosis may be as much as a centimeter in diameter. Peripheral infarcts reported in chickens have not been a common finding in turkeys. The kidneys are enlarged and as a rule slightly pale. The intestines appear anemic when superficially examined. There is always a marked catarrhal enteritis, and the contents are bile stained. The increase in urates is noted by the abnormal amount in the rectum; these are yellowish-green in color.

**Diagnosis.** An accurate diagnosis depends on finding spirochaetes in stained blood smears and tissues from typically sick individuals (Fig. 35.9). The organisms are readily stained by the Giemsa technic. Tunnicliff's technic for using her modified Gram's stain for spirochaetes is a simple and rapid method for staining blood smears for routine examinations.

**Prevention and control.** Since this disease is normally transmitted by such vectors as fowl ticks, mosquitoes, and fowl lice, a program for eradica-



Fig. 40.32. Spirochaetosis. Spleens from adult turkey hens, which show the typical mottling and ecchymosis. Approximately three-fourths normal size. (Univ. of Calif.)

tion of these will do much to prevent the spread of the disease. DDT has been found to be effective against fowl ticks. Drugs that have proven to be effective for the disease in chickens include the arsenicals (Morcos *et al.,* 1946) and penicillin (Nobrega and Bueno, 1945). Unpublished data from the University of California confirm the findings of Nobrega and Bueno and indicate that a single dose of 10,000 to 15,000 units of penicillin given intramuscularly to mature turkeys is highly effective as treatment if given when symptoms are first noted. The sulfonamides tried have not been effective.

Nobrega and Reis (1941) reported the successful use of a formalized vaccine prepared from infected chick embryos, for prevention of the disease. Their vaccine is prepared by inoculation of 12-day-old embryos with 0.05 to 1.0 cc. of chicken blood containing live spirochaetes. On the fifth day of incubation after infection, the organs of the embryo with the amniotic fluid are ground together and suspended in saline to make 30 cc. for each egg. Formalin is added to make 0.5 per cent, and the suspension is then left in the refrigerator for 24 hours before filtering through sterile gauze. This "stock suspension" is diluted with 3 parts of saline for use, and 1.0 cc. is injected intramuscularly in each bird.

## REFERENCES

Burroughs, A. L.: 1947. Fowl spirochetosis transmitted by *Argas persicus* (Oken), 1818 from Texas. Science 105:577.

Hinshaw, W. R., and McNeil, E.: 1946. Studies on a spirochaete found in the blood of sick turkeys. Jour. Bact. 51:599.

Hoffman, H. A., and Jackson, T. W.: 1946. Spirochetosis in turkeys. Jour. Am. Vet. Med. Assn. 109:481.

————, Jackson, T. W., and Rucker, J. C.: 1946. Spirochetosis of turkeys (a preliminary report). Jour. Am. Vet. Med. Assn. 108:329.

Hungerford, T. G., and Hart, L.: 1937. Fowl tick fever (spirochetosis), also transmitted by common red mite. Agr. Gaz. New So. Wales 48:591.

Kapur, H. R.: 1940. Transmission of spirochetosis through agents other than *Argas persicus.* Indian Jour. Vet. Sci. and An. Husb. 10:354.

Lesbouyries, G.: 1941. La Pathologie des Oiseaux. Vigot Frères, Paris, 868 pp.

Morcos, Z., Zaki, .O. A., and Zaki, R.: 1946. A concise investigation of fowl spirochetosis in Egypt. Jour. Am. Vet. Med. Assn. 109:112.

Nobrega, P., and Bueno, R. C.: 1945. A accao da penicilina na espiroquetose aviaria. Arq. Inst. Biol. São Paulo. 16:15.

———— and Reis, J.: 1941. Producao da vacina contra a espiroquetose aviaria em ovos embrionados. Arq. Inst. Biol. São Paulo. 12:87.

Reis, J., and Nobrega, P.: 1936. Tratado de Doencas das Aves. Instituto Biologico, São Paulo, Brazil.

Sreenivasan, M. K., and Sankaranarayan, N. S.: 1945. Spirochetosis of fowls in India. Indian Vet. Jour. 21:325.

Steinhaus, E. A., and Hughes, L. E.: 1947. Isolation of an unidentified spirochete from hen's eggs after inoculation with liver tissue from hens. U. S. Public Health Rpts. 62:309. (Reprint 2777.)

Stylianopoulos: 1925. (Original not seen, quoted by Lesbouyries (1941).)

van Heelsbergen, T.: 29. Handbuch der Geflügekrankheiten und der Geflügelzucht. Ferdinand Enke, Stuttgart, 608 pp.

Zuelzer, M.: 1936. Culex, a new vector of *Spirochaeta gallinarum.* Jour. Trop. Med. and Hyg. (London) 39:204.

## STAPHYLOCOCCOSIS

### (Staphylococcal Arthritis, Synovitis)

Jungherr (1933) described a disease characterized by arthritis. More recently Jungherr and Plastridge (1941) reported this disease and similar ones in poultry under the name staphylococcosis, caused by *Staphylococcus aureus* and *S. citreus.*

Madsen (1942) described a similar disease characterized by synovitis without involvement of the articular joints. Madsen's disease is probably the same as that described by Jungherr. It is becoming more and more prevalent in many turkey-growing areas, and may affect 2 to 10 per cent of the flock according to Madsen.

Symptoms vary with the acuteness of the disease. In the acute type, depression, decreased appetite, with watery sulfur-like droppings similar to those of blackhead, are common symptoms. Death may occur within 48 hours.

In less acute cases, the birds rest on their hocks, and show swollen joints. which are hot and painful to the touch (Fig. 40.33). The feet may be swollen and typical of the condition commonly called gout (Fig. 40.34 A and

B). Upon pressure the swelling fluctuates, indicating the presence of fluid exudate. In chronic cases, lameness is the chief symptom.

Autopsy of acute cases reveals an enlarged and dark liver, and congestion of the mucous membranes of the intestines. The intestinal contents are watery and yellowish in color. Inflammation of the synovial membranes of the hock joints with increased fluid is characteristic. Chronic cases show principally involvement of the joints and muscles of the legs and feet. The exudate may vary from a semigelatinous to a cheeselike flaky consistency.



FIG. 40.33. Staphylococcal arthritis in young poult. (Hinshaw, Univ. of Calif.)

No remedy is known. Madsen tried sulfapyridine without success. Trials made at the University of California (unpublished) show little promise for the sulfonamides or for penicillin against this disease. The general recommendations for handling other infectious diseases are suggested.



A.      B.
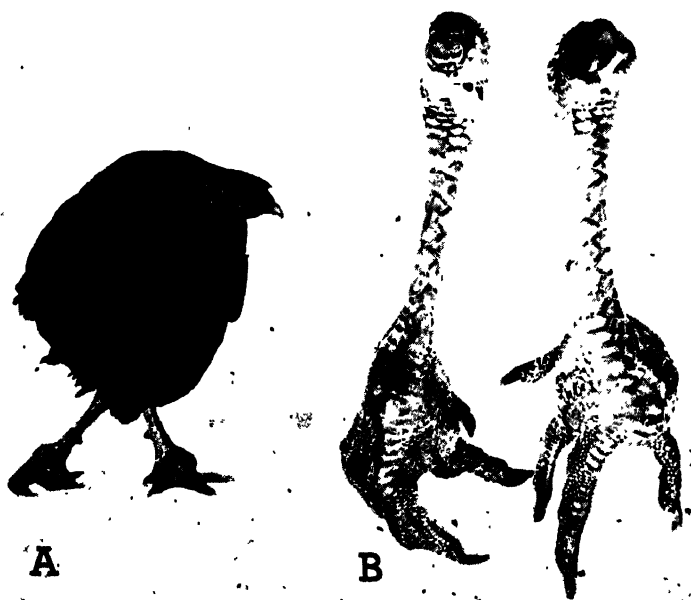
FIG. 40.34. A—staphylococcal arthritis in an adult turkey. Note swollen joints of the feet. B—close-up of the feet of the turkey shown in A. (Hinshaw, Univ. of Calif.)

## REFERENCES

Jungherr, E.: 1933. Staphylococcal arthritis in turkeys. Jour. Am. Vet. Med. Assn. 82:243.
———— and Plastridge, W. N.: 1941. Avian staphylococcosis. Jour Am. Vet. Med. Assn. 98:27.
Madsen, D. E.: 1942. Synovitis of turkeys. Turkey World 17 (2) :24.

## STREPTOCOCCOSIS

Generalized infections in turkeys caused by streptococci are being diagnosed with increasing frequency. Volkmar (1932), reporting several outbreaks of apoplectiform septicemia in turkeys caused by a streptococcus, described the disease as resembling fowl cholera. The losses are sporadic in nature, the disease is very acute, and symptoms are seldom seen before death. The principal lesions noted on autopsy are congestion or diffuse hemorrhages in the skin and breast muscles, together with generalized congestion of the internal organs. Hemorrhagic enteritis and peritonitis are common, and the heart sac may be filled with a blood-tinged fluid. The disease must be differentiated by bacteriologic studies.

Acute outbreaks of a disease in young poults with symptoms and autopsy findings resembling pullorum disease have been encountered by us. Losses in these cases have equalled those of pullorum disease or paratyphoid infections. Examples of mortality experienced in these outbreaks are 430 out of 1,080 poults; 300 out of 1,200 poults; and 450 out of 1,500 poults. Losses in these outbreaks started within the first week and continued for two weeks. The symptoms resembled those of pullorum disease. Necropsy findings included congestion and necrosis of the lungs, congestion and necrosis of the liver, and enteritis. Pin-point areas of necrosis in the livers were especially common. A short-chain streptococcus which has the characteristics of *Streptococcus zymogenes* has been consistently isolated from these cases. No detailed studies have been made of this disease, but mention is made of it since it may be confused with pullorum disease.

## REFERENCE

Volkmar, F.: 1932. Apoplectiform septicemia in turkeys. Poultry Sci. 11:297.

## TUBERCULOSIS

Tuberculosis, a chronic disease affecting turkeys and other fowls, is caused by *Mycobacterium avium* Chester. It is not common in commercial turkey flocks, and all the outbreaks studied at this station (California) have been associated with tuberculous chickens.

**Symptoms.** There are no typical symptoms. Lameness and emaciation have occasionally been observed. Many turkeys that show lesions on autopsy maintained their weight for several months before death. Tuberculous turkeys placed in individual cages and observed for periods of one to ten weeks held their initial weight, and a few even gained. Such birds often go

through intermittent periods of normality and depression lasting for two or three weeks before death. In the periods of depression, which last for 2 or 3 days, the feathers become ruffled, the appetite diminishes, and diarrhea develops. These periods are followed by a few days of normal appetite and general improvement of health.

**Clinical diagnosis.** Tuberculosis in turkey flocks has, in the experience of the writer, been more often detected by accidental discovery of lesions during an autopsy by the owner, or by the housewife while preparing a bird for roasting, than by symptoms seen in the flock or by the use of the tuberculin test. Hinshaw, Niemann, and Busic (1932) found that about 75 per cent efficiency can be expected from the use of the tuberculin test as a means of diagnosing tuberculosis in turkeys. The edge of the wing web proved to be the best site for inoculation of the tuberculin, but the results, even in this area, were more difficult to interpret than in other animals.

**Autopsy findings.** The gross pathology of tuberculosis in turkeys has not been found markedly different from that of the disease in chickens. The distribution of lesions in turkeys indicates a tendency for a greater number of organs to become infected than in chickens, and as in chickens, the disease is principally abdominal in nature. Seven cases of tuberculosis in turkeys from five California outbreaks have been typed and found to be of avian origin.

A study of the distribution of lesions in turkeys from seven California outbreaks showed that the liver, bone marrow, spleen, intestines, ovaries, mesentery, skin, thymus gland, and lungs were, in the order given, the most common seats of lesions. The ovary and the thymus glands were more often found to be infected than in chickens. Attention is also called to the large percentage of cases of bone-marrow lesions. The number of birds examined for bone-marrow lesions was small as compared to the total, but they were in all stages of the disease. When lesions were found in the bone marrow, they were always found in at least one other organ.

**Differential diagnosis.** Some of the conditions noted in turkeys which might be confused with tuberculosis are mycosis, blackhead, and tumors. Mycotic lesions in the liver and kidney, which on first glance are suggestive of tubercles, have been observed. These are not definitely encapsulated and circumscribed, however. On microscopic examination, mycelia are found, while acid-fast rods cannot be demonstrated.

Blackhead, or infectious enterohepatitis, should not be confused with tuberculosis, because the lesions in the liver do not resemble tubercles. Furthermore, the well-known characteristic lesions of the disease in the ceca should help to differentiate it from tuberculosis. On the other hand, tumors of the liver and ovary have been noted that were suggestive of tuberculosis until a microscopic examination was made.

**Prevention, control, and treatment.** Complete isolation of turkeys from

chickens will do much to prevent tuberculosis. Once the disease is found, it is a good plan to dispose of the entire flock, as well as all chickens on the premises. The best way to dispose of a tuberculous flock of turkeys or chickens is to sell them subject to condemnation. By such a scheme all birds showing lesions of tuberculosis on drawing are destroyed, and the owner is paid for the ones that are suitable for food.

Day-old poults, rather than adult stock, should be purchased as replacements. They should be brooded away from the infected area and should not be allowed to range there for at least one year after the diseased birds have been disposed of. A careful grower may sell the entire flock each year and start with day-old stock each spring for several years in order to insure freedom from tuberculosis.

## REFERENCE

Hinshaw, W. R., Niemann, K. W., and Busic, W. H.: 1932. Studies of tuberculosis of turkeys. Jour. Am. Vet. Med. Assn. 80:765.

## MISCELLANEOUS BACTERIAL DISEASES

Numerous reports of occasional losses due to miscellaneous bacteria have been made by investigators. These include the reports of Stafseth (1939) and Stafseth, Mack, and Ryff (1940) on Pseudomonas infections.

In one outbreak caused by a hemolytic Pseudomonas, Stafseth states that there was a morbidity rate of 50 per cent, and a low mortality. The outstanding autopsy findings are: very dark and often uncoagulated blood, pinpoint areas of necrosis or yellowish-gray streaks found in the liver, mottled spleen, and hemorrhagic enteritis. According to these investigators the species of Pseudomonas responsible for the outbreak in turkeys differs in several respects from *P. aeruginosa,* a common inhabitant in fowls.

Frequently, diagnostic laboratories report the isolation of organisms belonging to the colon-aerogenes group of bacteria from outbreaks of septicemic diseases in young poults. In similar outbreaks organisms belonging to the paracolon groups are also reported. Paracolon types belonging to the Arizona group of Edwards, West, and Bruner (1947) cause symptoms and mortality similar to those seen in salmonellosis. Two of these types which have caused heavy losses in turkey poults have been described by Hinshaw and McNeil (1944, 1946). Both of these have been shown to be hatchery transmitted and to be carried by snakes in the same manner as salmonellosis.

Such outbreaks should be handled in the same manner as those due to other infections. Sanitation and management play large parts in their prevention and control.

*Proteus* infections occasionally occur in young poults and may account for severe losses. Such infections are usually secondary to some environmental setback.

Fenstermacher and Pomeroy (1939) described losses in breeding turkey

hens associated with several species of Clostridium. The outbreak involved a flock of 1,000 turkey hens in which there was a low mortality. The losses occurred during the early part of the breeding season, and the investigators state that the atria of infection were probably wounds inflicted at the time of mating. Subcutaneous edema and emphysema around the head and thigh were the principal symptoms reported. Injuries due to mating can be prevented by proper management. (See section on injuries.)

## REFERENCES

Edwards, P. R., West, M. G., and Bruner, D. W.: 1947. Arizona group of paracolon bacteria. A new group of bacteria pathogenic for animals and probably also for man. Ky. Agr. Exper. Sta., Bul. 499:1.

Fenstermacher, R., and Pomeroy, B. S.: 1939. Clostridium infection in turkeys. Cornell Vet. 29:25.

Hinshaw, W. R., and McNeil, E.: 1944. Gopher snakes as carriers of salmonellosis and paracolon infections. Cornell Vet. 34:248.

———— and McNeil, E.: 1946. The occurrence of type 10 paracolon in turkeys. Jour. Bact. 51:281.

Stafseth, H. J.: 1939. Pseudomonas infection in turkeys. Poultry Sci. 18:412.

————, Mack, W., and Ryff, J. F.: 1940. Pseudomonas infection in turkeys. Poultry Sci. 19:126.

## PROTOZOAN DISEASES[3]

### BLACKHEAD

#### (Infectious Enterohepatitis)

Blackhead, first described by Cushman (1893), is caused by a protozoan parasite, *Histomonas meleagridis.* It is credited with being the cause of the temporary abandonment of the turkey industry in some sections of eastern and midwestern United States. The early researches of Smith (1895), Moore (1896), Curtice (1907), Higgins (1915), Smith and Graybill (1920), and Tyzzer (1920) paved the way for later studies which proved that the disease is preventable.

**Etiology.** *Histomonas meleagridis,* the causative agent, is classified as a flagellate but is one of the few that likewise has an amoeboid stage. It is harbored by the common poultry cecal worm, *Heterakis gallinae,* found in the ceca, or blind pouches, of a large percentage of chickens. This, together with the fact that chickens are not, as a rule, highly susceptible to the parasite, has frequently been responsible for the transmission of the disease from apparently healthy chickens to turkeys. The most recent paper on the morphology of *Histomonas meleagridis* is that of Wenrich (1943).

The parasites are capable of living for long periods in the cecal worm and its eggs. Van Es and Olney (1934) found that the infection remained on vacant yards from the middle of November until the middle of June during each of five years when turkeys were reared in the yards from June to November. For a more detailed discussion of the parasite and a description of the organism, reference is made to the works of Tyzzer (1920), Delaplane (1932), and DeVolt and Davis (1936). (See also chapter on Protozoa.)

**Symptoms.** "Blackhead," the common name for infectious enterohepatitis, is a misnomer. Sometimes the head does become darkened, but this symptom is not characteristic of blackhead alone. Drowsiness, weakness, drooping wings and tail, a lowered head, ruffled feathers, and constant sulfur-colored diarrhea are characteristic symptoms. As a rule, adult birds are sick for several days before dying and become very emaciated. Young poults may have a very acute type of the disease and may die soon after symptoms are noted. Although turkeys of all ages are susceptible, the heaviest losses occur during the first twelve weeks of life. Another peak of mortality is often observed after the birds are put on the finishing ration to prepare them for market. Sometimes a third peak of losses occurs during the breeding season.

The mortality is high, often approaching 100 per cent of the flock, and averages about 50 per cent unless kept under control. Once the disease attacks a flock, occasional birds are likely to die between the intermittent periods of heavier losses, especially if the flock is not moved frequently to clean grounds. The period of incubation after contact with infection is 15 to 21 days.

**Autopsy findings.** The liver and the ceca are the principal organs showing marked changes



FIG. 40.35. Ceca of a turkey affected with blackhead. Note the swollen condition of one cecum, and the discolored diseased areas near the middle and at the tip. The other shows a single lesion near its middle portion. (Graybill, Univ. of Calif.)

caused by blackhead. The severity of these changes varies with individuals. The cecal lesions are apparently the primary ones, and one or both ceca may be affected (Fig. 40.35). The lesions consist of marked inflammatory changes and ulcerations, sometimes involving most of the organ. A

single ulcer may involve the serosa and form an opening through the entire wall. The mucous membrane often becomes necrotic, much thickened, and covered with a characteristic foul-smelling, yellowish-green, semi-caseous exudate, or a dry, hard, cheesy core may fill the cecum.

The affected liver (Fig. 40.36) presents a characteristic appearance, with



FIG. 40.36. Liver of turkey affected with blackhead. (Graybill, Univ. of Calif.)

areas of necrotic and degenerated tissues on the surface. These are more or less circular, have a yellowish to yellowish-green appearance, and in contrast to tumors and tubercles (tuberculosis), are somewhat depressed below the liver surface. They extend deeply into the tissue and are more or less con-fluent with the healthy tissue. In older birds the individual lesions are often merged. Evidence of healing is seen in the large amount of scar tissue in older birds. Occasionally, peritonitis and involvement of the other organs may be observed.

Differential diagnosis. Blackhead must be differentiated from other diseases involving the liver and cecum. Chief among these are tuberculosis, tumors, and mycotic diseases. Allen (1941) has described a disease of the liver which she claims to be due to a species of Trichomonas. The lesions in this disease are easily differentiated from those of blackhead, as are the lesions of tuberculosis and mycotic infections of the liver. Demonstration of the causative agent by microscopic examination and culturing are necessary for final diagnosis.

**Treatment.** No drug or combination of drugs has been found entirely satisfactory for stopping losses from blackhead once the disease has appeared in a flock. Nicotine products and phenothiazine, often falsely called blackhead remedies, have gained their reputation because of claimed successes in removal of cecal worms, thus aiding prevention of the disease. Neither agent has been proven effective against *Histomonas meleagridis*, the causative agent of blackhead.

Mapharsen (meta-amino-parahydroxy-phenylarsine hydrochloride), a drug containing 29 per cent of arsenic in trivalent form, has been reported by Blount (1938a, 1938b), Bolin and Vardiman (1941), and McCulloch and Nicholson (1941) as a promising remedy for treatment of blackhead. The trials reported by all these investigators concern small numbers of turkeys, and more data are necessary before the drug can be considered a specific remedy for this disease. The average dose for a 9-pound turkey given by Bolin and Vardiman was 6 milligrams of mapharsen dissolved in sterile distilled water (amount not given) and injected intramuscularly. McCulloch and Nicholson gave each of twenty-seven sick turkeys weighing from 8 to 12 pounds, 5 milligrams dissolved in 1.0 cc. of sterile water intramuscularly (pectoral muscles). According to them, seventeen (63 per cent) made complete recovery.

**Prevention.** Blackhead is a "filth-borne" disease dependent on carriers, including not only chickens and turkeys but other birds as well. For a discussion of carriers other than chickens and turkeys the reader is referred to the chapter on Protozoa. These carriers eliminated the causal organism in the feces, alone or within the cecal worm and its eggs. When the organism is ingested by susceptible stock, infection results. Because there is no practicable method of identifying carriers, all chickens and turkeys must be under suspicion. Examples of methods of mechanical transmission of the disease are feed sacks, and grains (corn, oats, wheat, etc.) that become contaminated with feces from chickens or turkeys that harbor both the cecal worm and the blackhead parasite.

The need for prevention is greatest during the most susceptible age, from hatching to twelve weeks. Van Es and Olney (1934) suggest the following requirements for preventing losses from blackhead:

1. Artificial incubation in order to escape the hazard arising from close association with the parent bird in the same environment.

2. Brooding in an enclosure from which all infection hazards have been previously excluded by attention to such details as hardware-cloth floor covering, and all other measures by which actual contact with soil can be avoided.

3. Maintenance of the poults, at least up to twelve weeks old, on clean ground not previously occupied by either turkeys or chickens.

4. Provision of a wide range for the maturing bird—if possible, one not

previously occupied by blackhead-infected fowl. If such an environment is not available and the turkeys must be confined in more constantly occupied enclosures, yards should be covered either with coarse gravel or with 1-inch hardware cloth.

5. Maximum protection against the fecal contamination of food and water by the use of feeding and watering equipment specially designed for the purpose (see pp. 93 to 97).

Billings (1928) modifies this method of rearing turkeys free from blackhead. The principal difference in the two plans is in the substitution of a four-yard rearing system by Billings for the fourth step in the Van Es and Olney plan. The four-yard rearing system consists of dividing 1 acre into four yards. These are divided as suggested in Figure 40.37. Three hundred poults can be raised in such a unit. The poults are reared for a month in each of the other yards in succession. They are moved each month until marketed. The acre of ground can be fenced into the 1/4-acre sections, or the fence may be a temporary one, set up around a different section each month.
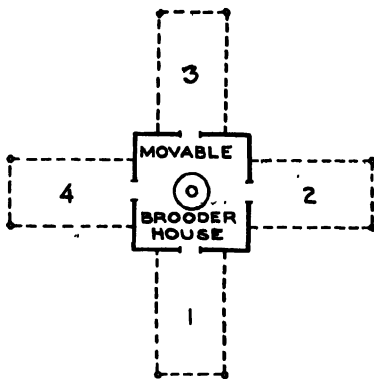
Regardless of the system used in rearing turkeys, the following precautions against blackhead must also be observed:

1. Keep the turkeys entirely separated from the chickens or chicken yards. Drainage from chicken yards to turkey yards is a common source of blackhead.

2. Do not rear turkeys on ground that has been fertilized with chicken or turkey manure.

3. Do not rear turkeys in yards where losses from blackhead have occurred until at least one year has passed after the removal of the last diseased bird.
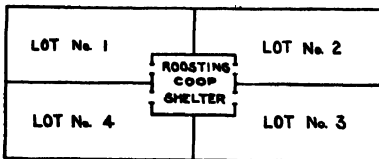


FIG. 40.37. Two systems for rotation of runs suggested by Billings for prevention of blackhead. (Hinshaw, Univ. of Calif.)

4. Do not introduce new stock without quarantining it for three weeks before adding it to the flock.

5. Feed an adequate ration, with plenty of fresh, clean water.

The continual feeding of tobacco dust mixed with the mash as a preventive has been suggested by several experiment station investigators. The principle of this plan is to prevent cecal worms from becoming established in the flock and thus to reduce the chances of transmission of the blackhead

parasite by this means. Scott (1935) recommends adding 4 pounds of tobacco dust containing at least 2 per cent nicotine to each 100 pounds of mash; this mixture is to be fed continuously from the time the poults are transferred from the brooders to the range.

McCulloch and Nicholson (1940), and Nicholson and McCulloch (1941) reported phenothiazine as an effective remedy against the cecal worm and suggest its use in the prevention of blackhead. This drug is relatively nontoxic for chickens and turkeys and may prove of value as a preventive in areas or on ranches where the cecal worm is very prevalent and blackhead cannot be controlled in any other way. The dosage recommended for turkey poults is ½ gram per poult and for adults 1 gram per bird. Van Ness and Hamilton (1944) and Wehr and Olivier (1946) have shown that the drug is not effective in preventing the disease on heavily infected ground even when fed continually in the mash for extended periods. Wehr and Olivier fed as high as 2 per cent phenothiazine in mash for as long as six weeks without significantly lowering the incidence of the disease. An important reason for this failure is that *Histomonas meleagridis* can be directly transmitted from bird to bird without the aid of the cecal worm. (For a more complete discussion of direct transmission see chapter on Protozoa.)

*Neither tobacco dust nor phenothiazine are remedies against blackhead itself and should not be used as such. Such remedial measures as just described are not recommended for general use where blackhead can be readily controlled by methods not involving drugs. Nor are these remedies recommended as general procedures if blackhead has not been proven a problem.*

## REFERENCES

Allen, E. A.: 1941. Macroscopic differentiation of lesions of histomoniasis and trichomoniasis in turkeys. Am. Jour. Vet. Res. 2:214.

Billings, W. A.: 1928. Talking turkey. Minn. Agr. Ext. Div., Spec. Bul. 124.

Bolin, F. M., and Vardiman, P. H.: 1941. Mapharsen as a treatment for enterohepatitis of turkeys. Jour. Am. Vet. Med. Assn. 98:229.

Blount, W. P.: 1938a. Clinical notes on poultry diseases. Vet. Jour. 91:278.

———: 1938b. New arsenical preparations in the treatment of blackhead in turkeys. Vet. Jour. 94:344.

Curtice, C.: 1907. The rearing and management of turkeys with special reference to the "blackhead" disease. R. I. Agr. Exper. Sta., Bul. 123.

Cushman, S.: 1893. Experiments with turkeys. R. I. Agr. Exper. Sta., Rep. for 1893, p. 284.

Delaplane, J. P.: 1932. Etiological studies of blackhead (enterohepatitis) in turkeys. R. I. Agr. Exper. Sta., Bul. 233.

DeVolt, H. M., and Davis, C. R.: 1936. Blackhead (infectious enterohepatitis) in turkeys, with notes on other intestinal protozoa. Md. Agr. Exper. Sta., Bul. 392:493.

Higgins, C. H.: 1915. Entero-hepatitis or black-head in turkeys. Canad. Dept. Agr., Health Animals Branch, Bul. 17.

McCulloch, E. C., and Nicholson, L. G.: 1940. Phenothiazine for the removal of *Heterakis gallinae* from chickens. Vet. Med. 35:398.

——— and Nicholson, L. G.: 1941. Mapharsen therapy in enterohepatitis of turkeys. Vet. Med. 36:574.

Moore, V. A.: 1896. The direct transmission of infectious entero-hepatitis in turkeys. U.S.D.A., Bur. An. Ind., Cir. 5:1.

Nicholson, L. G., and McCulloch, E.: 1941. New drug shows promise for controlling blackhead. Turkey World 16:12, 13, and 48.

Scott, H. M.: 1935. Kansas controls blackhead by feeding tobacco dust. Turkey World 10:10, 12, and 48.

Smith, T.: 1895. An infectious disease among turkeys caused by protozoa (infectious enterohepatitis). U.S.D.A., Bur. An. Ind., Bul. 8:7.

―――― and Graybill, H. W.: 1920. Blackhead in chickens and its experimental production by feeding embryonated eggs of *Heterakis papillosa*. Jour. Exper. Med. 32:143.

Tyzzer, E. E.: 1920. The flagellate character and reclassification of the parasite producing "Blackhead" in turkeys—*Histomonas* (Gen. Nov.) *meleagridis* (Smith). Jour. Parasit. 6:124.

Van Es, L., and Olney, J. F.: 1934. Diseases of poultry—their nature and control. Nebr. Agr. Exper. Sta., Bul. 290.

Van Ness, G., and Hamilton, C. M.: 1944. The use of phenothiazine in the control of enterohepatitis of turkeys. 54th Ann. Rep. Wash. Agr. Exper. Sta., Bul. 455:152.

Wehr, E. E., and Olivier, L. G.: 1946. Limitations of phenothiazine in the control of cecal worms and blackhead disease of turkeys. Poultry Sci. 25:199.

Wenrich, D. H.: 1943. Observations on the morphology of Histomonas (Protozoa, Mastigophora) from pheasants and chickens. Jour. Morph. 72:279.

## COCCIDIOSIS

Losses in young turkeys are often mistakenly ascribed to coccidiosis when some other disease is responsible. Most of the cases of bloody diarrhea in turkeys which are attributed to coccidia by some observers are not caused by these parasites.

Only two species of turkey coccidia have been described: *Eimeria meleagridis,* and *E. meleagrimitis.* Neither species is pathogenic for chickens, and none of the seven species of coccidia described for chickens has been definitely shown to be pathogenic for turkeys. For a complete description of these organisms the reader is referred to Tyzzer (1929) and Becker (1934).

**Symptoms and mortality.** The symptoms of coccidiosis in turkeys differ considerably from those often seen in acute outbreaks of the disease in chicks. When trying to diagnose coccidiosis, therefore, one should not attempt to compare the symptoms of turkeys with those seen in chicks. The presence of the disease is strongly suggested by listlessness, drooping wings, ruffled feathers, and a light brownish diarrhea with considerable mucus.

Under good management and sanitary conditions, coccidiosis does not cause severe mortality in turkeys. Although heavy losses have been associated with the disease, some contributing factor, such as insanitary surroundings or inadequate diets, has been observed in the outbreaks studied.

**Autopsy findings.** Catarrhal enteritis especially in the lower half of the intestine is characteristic. In marked cases the intestine may be filled with whitish-gray semigelatinous pus containing myriads of coccidia. This exudate adheres to the intestinal wall and leaves a denuded area when scraped from the surface. Only by microscopic examination can coccidiosis be definitely diagnosed.

**Transmission.** A large percentage of the poults that survive an outbreak continue to be carriers and shed the parasites in their droppings. Thus, as

long as adult turkeys are kept on the premises while poults are being brooded, the adults are a continual source of infection. The means of transmission to poults in the brooder are many and include flies, tools, feed sacks, feed, and attendants.

Hinshaw (1937) reported two experiments designed to show how coccidiosis may be mechanically transmitted. One of these, conducted to determine how far coccidia can be carried by a person visiting an infected yard, proved by repeated trials that coccidial oocysts can be carried as far as ½ mile on the soles of shoes and still remain capable of sporulating and producing disease. The second experiment was conducted to determine the possibility of transmitting coccidiosis by contaminated feed. Sterilized feed that was walked on by attendants who had previously visited infected yards, produced coccidiosis when fed to susceptible birds.

**Prevention, control, and treatment.** Prevention of coccidiosis in turkeys is best accomplished by preventing contact of the poults with the adult stock.

In artificial brooding, preventive measures are more practicable than when turkeys are brooded naturally; but two important avenues of infection exist—namely, the feed and the attendant. Indirectly, the attendant is a carrier of coccidia by way of feed, especially if he shovels it from one pile to another on a floor, since he cannot avoid walking on feed mixed in this manner. Visitors are also potential mechanical carriers.

Buying day-old poults from reliable hatcheries and using artificial brooding methods are recommended for the turkey grower who has a heavily infected flock of adult turkeys. In such instances all adult stock should be disposed of several weeks before the poults are purchased, and the poults should be reared in houses and yards that have not been used for the adults. This procedure eliminates the most important source of infection, the adult turkey.

Coccidial oocysts must have moisture in order to form spores, without which they cannot produce disease. Keeping thoroughly dry all areas to which poults have access will do much, therefore, to prevent acute outbreaks. Frequent changing of litter, the use of wire-screened platforms for water and feed containers, and ample floor space, are aids in keeping the floors of the brooder houses dry. The methods suggested for preventing blackhead will also aid in preventing coccidiosis.

Control of the disease in acute outbreaks may be accomplished by rigorous dry cleaning at daily intervals. Feeding a 40 per cent dried-milk mash may aid in the control program, but greater effort must be made to keep the floors dry while the milk treatment is used. A laboratory diagnosis must be obtained before this treatment is begun, because the treatment may cause severe losses in some types of enteritis not due to coccidiosis. It should con-

tinue only while justified by the response of the birds and should stop immediately if heavy losses are experienced.

The sulfonamides have proven of value in control of coccidiosis of chickens, but there is no literature available on their use for turkeys. Field trials have indicated that they are worthy of trial in acute outbreaks. The reader is referred to the section on chicken coccidiosis for reference to methods of treatment. In most outbreaks good results will be obtained by following the above precautions without treatment.

### REFERENCES

Becker, E. R.: 1934. Coccidia and Coccidiosis of Domesticated, Game and Laboratory Animals, and of Man. The Iowa State College Press, Ames, Ia.
Hinshaw, W. R.: 1937. Diseases of turkeys. Calif. Agr. Exper. Sta., Bul. 613.
Tyzzer, E. E.: 1929. Coccidiosis in gallinaceous birds. Am. Jour. Hyg. 10:269.

### HEXAMITIASIS
#### (Infectious Catarrhal Enteritis)

Hexamitiasis is a disease of young poults, causing its greatest mortality in those under ten weeks of age. It was thought for many years that the disease was caused by trichomonads, but Hinshaw, McNeil, and Kofoid (1938a, 1938b) reported that a species of Hexamita and not a Trichomonas was responsible for the disease. They found that neither *T. eberthi* nor *T. gallinarum* (commonly found in the ceca of turkeys and chickens) was capable of producing a similar disease. The causal agent has been named *Hexamita meleagridis* by McNeil, Hinshaw, and Kofoid (1941). They describe the parasite as follows:

"This organism (minus flagella) varies in length from 6 to 12μ (average 9μ) and in width from 2 to 5μ (average 3μ). The nuclear membrane is distinct, and the karyosomes are round and fairly large (two-thirds diameter of the nucleus). Anterior to the nuclei are 2 large blepharoplasts (or groups of blepharoplasts) from which arise the 4 anterior and 2 anterolateral flagella. The flagella are all of about the same length, measured from the point of emergence from the body. The 4 anterior flagella are usually curved back along the body. Just posterior to these 2 large blepharoplasts are 2 others from which arise the 2 caudal flagella. These flagella pass posteriorly in a granular line of cytoplasm to their pockets of emergence near the posterior end of the body" (Fig. 40.38 A and B).

Unpublished reports received by Hinshaw indicate that the disease is prevalent in most sections of United States. Published reports in addition to those from California include one from Connecticut by Jungherr and Gifford (1944) and one from Indiana by Doyle, Cable, and Moses (1947). Campbell (1945) reported the presence of Hexamita in turkeys infected also with Cochlosoma, in an outbreak in Scotland. He considered that Cochlosoma was the primary cause of losses in this outbreak.

**Symptoms.** In the early stages of acute outbreaks, the poults are nervous and require more heat than normally; the body temperature is normal or subnormal; the gait is stilted and the feathers are ruffled and unkempt. There is foamy watery diarrhea, but the cecal droppings do not appear changed.

In most cases the poults continue to eat and may even appear to consume more feed due to a nervousness that is always evident. This nervousness is also manifested by the continual chirping of the birds especially in the early stages. Due to improper digestion and assimilation of feed, the poults lose
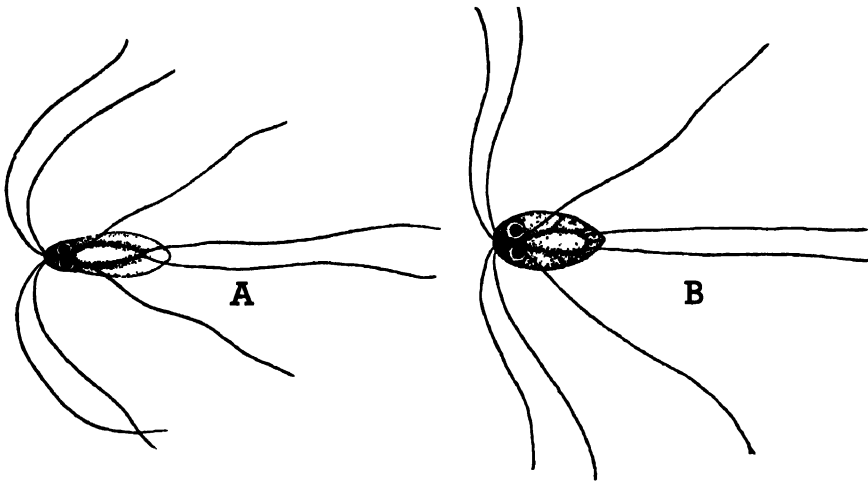


FIG. 40.38. A, B—*Hexamita meleagridis* from the intestine of the turkey, showing individual variation in size and shape. ×1,875. (McNeil, Hinshaw, and Kofoid, Am. Jour. Hyg.)

weight rapidly. Many of the survivors continue to be underweight for weeks. In the later stages of the disease the poults become listless, sit under the hover, and go into a coma. Finally, they struggle, flap their wings, and die.

Subacute attacks of the disease may occur in young poults early in the season before the infection reaches the epidemic stage. Milder outbreaks may occur in poults that reach a resistant age. In such outbreaks, listlessness and loss of weight are the most prominent symptoms. Loss of appetite will depend on the severity of the disease. Large numbers of stunted individuals result from this form.

**Course and mortality.** In experimentally produced outbreaks symptoms appear in 4 to 7 days after ingestion of the parasites. The period of incubation varies with the amount of inoculum and the age of the individual. If temperatures are taken daily following infection, a drop will often be noted a day before visible symptoms are seen. Likewise, daily weight records will usually show a decline in the daily gain before symptoms appear.

Mortality may start within a day after symptoms appear. In acute out-

breaks the course of the disease is typical of an acute infectious disease with the peak of mortality occurring in 7 to 10 days following the appearance of symptoms. In most instances straggling losses occur for as long as three weeks, and in a few flocks, a second peak of mortality has been observed.

In outbreaks complicated with other infections, such as salmonellosis, or by faulty management, the course may be varied and the mortality increased. Heavy losses seldom occur in poults over ten weeks of age unless there has been some lowering of resistance due to another infection or due to environmental factors. It is always desirable to eliminate the possibility of such complications whenever losses associated with the presence of *Hexamita meleagridis* are encountered in older turkeys.

Environmental and husbandry factors as well as age greatly influence the mortality. It may vary from a few poults to the entire flock. Under experimental conditions, using normal poults, we have not been able to produce heavy mortality in poults over eight weeks of age.

**Autopsy findings.** At autopsy the birds are in poor condition; the feathers lack luster; the skin is dry; the flesh of the breast is dehydrated and reddened. In young poults suffering from an acute outbreak the crop usually contains some food; in poults that linger longer before death it is usually empty.

The principal pathological change occurs in the upper intestine (duodenum, jejunum, and ileum), where there is catarrhal inflammation with marked lack of tone. The intestinal contents may vary from a thick mucous type to a thin watery, foamy type. The latter is most characteristic. Localized bulbous areas filled with watery contents are also characteristic. The mucous membrane of these areas is often congested.

The contents of the ceca may be more fluid than normal, but seldom changed otherwise. The only pathology noted in the ceca is a congestion of the cecal tonsils.

**Diagnosis.** Diagnosis must be based on finding Hexamita in the upper intestines. The examination of cecal or rectal contents is not recommended as a diagnostic procedure because of the necessity of having to differentiate other flagellates from *Hexamita meleagridis* (Fig. 40.38). If smears are made from the duodenum or jejunum, and diluted with physiological saline, Hexamita, if present, will usually be found free from other flagellates. Cochlosoma, a flagellate first reported in ducks by Kimura (1934) has been found by McNeil and Hinshaw (1942) associated with *Hexamita meleagridis* in a few outbreaks. Cochlosoma can be readily distinguished from Hexamita by its characteristic rolling movement and its cochleal shape. It is necessary for best results to examine poults that have recently died, but the parasites have been found in refrigerated specimens 48 hours after death. It may be

necessary to warm the slide at 35–40° C. for a few minutes, if the smears are taken from such specimens.

The parasites may also be found in the bursa of Fabricius; in the carrier stage they localize in this organ as well as in the cecal tonsils. When the acute disease subsides, Hexamita can seldom be found in the upper intestines.

**Transmission.** McNeil, Platt, and Hinshaw (1939) found *Hexamita meleagridis* in California valley quail, Gambel's quail, and in chukar partridges. Hinshaw and McNeil (1941), in a survey of possible carriers, found Hexamita in 16.5 per cent of seventy-nine live adult turkeys by rectal examination, and in 32.4 per cent of seventy-four turkeys by examination of scrapings from the cecal tonsil at necropsy. All these birds were survivors of acute outbreaks. They also examined eleven species of game and wild birds other than quail and chukars killed on or near infected ranches. None harbored Hexamita. Chickens, ducks, pigeons, and guinea fowl were negative for *Hexamita meleagridis,* but pigeons harbored *Hexamita columbae* (McNeil and Hinshaw, 1941a). Ducks and chickens were artificially infected with *Hexamita meleagridis,* but symptoms of the disease were not produced. Kimura (1934) in a paper on Cochlosoma in ducks mentions the occurrence of Hexamita in the ceca and large intestine of domesticated ducks in California. McNeil and Hinshaw (1941b) found the parasite in chickens artificially infected twenty-two weeks after inoculation. Hexamita has also been reported in peafowl (California State Department of Agriculture, 1941) and in pheasants by Hinshaw and McNeil (1942) and Stover (1943). Thus, surviving turkeys, quail, chukars, pheasants, peafowl, chickens, and ducks must be considered potential carriers of *Hexamita meleagridis.*

No insect transmitter has been found. It has consistently been possible to keep noninfected poults free from the disease when reared in proximity to infected brooders even though flies were abundant.

**Epidemiology.** Management and environment play important roles in the transmission of this disease. The adult turkey that has survived an outbreak is the most important factor in perpetuation of the disease on a ranch. Chukar partridges, quail, pheasants, peafowl, and ducks may also play a part in starting an outbreak. Even though chickens have not been found infected under natural conditions, the fact that they may be artificially infected means that the parasite may sometimes be adapted to them, and they must be considered potential carriers. Outbreaks of hexamitiasis have been definitely traced to infected quail and pheasants ranging with turkeys.

In the field the acute disease is usually seen in the later hatches, and often after the breeders have been marketed. Thus adult transmission is often difficult for the owner to understand, because his earlier groups did not show symptoms. The explanation based on experimental and field studies is that

the early hatched poults act as a reservoir for increasing the dosage and prob-
ably the virulence of the parasites, and serve as the intermediary transmitter
from the breeders to the later hatched poults. Experimentally, it takes from
three to five passages of parasites removed from turkeys in the carrier stage
through young poults before the infection is increased to insure acute out-
breaks.

Environmental and husbandry factors also play an important part in
establishing the infection in epidemic proportions. In most instances, as the
brooding season advances, the volume of work on the ranch increases, and
the space available per bird is less, thus increasing the chances for spread of
disease. The difficulties arising with the hot weather, and in some areas late
spring fogs, also contribute to the development of the disease in later broods.

**Prevention, control, and treatment.** The primary source of infection is
the intestinal contents of carriers. The entire program of prevention must be
built around the recognition of this fact. Finding a satisfactory method of
preventing the transfer of droppings from carriers to young birds is the most
efficient method of preventing disease. *No general recommendation as to the
best procedure to follow can be given because every ranch requires a separate
solution of the problem of eliminating the danger of having carriers on the
ranch.*

Factors which will aid in solving the individual problems are:

1. Separate units and caretakers for the breeding flock and the young
poults.
2. Separate equipment for each age group.
3. Intelligent use of wire platforms for feed and water.
4. Intelligent use of cement yards and wire pens.
5. Arrangement of feeding and watering equipment for easy access to
the attendant without entering the pen, to avoid contamination.
6. If the poults have undergone an outbreak of pullorum disease or
paratyphoid infection, avoid changes in brooding until they are twelve to
sixteen weeks of age.
7. Sell all breeding birds two weeks before any poults are hatched.
8. Avoid ranges frequented by pheasants, quail, and chukars.

An accurate laboratory diagnosis is the first essential in the advent of a
suspected outbreak. Live sick birds are necessary for the accurate diagnosis
of hexamitiasis, although Hexamita may be found as long as 48 hours after
death of the poult if decomposition has not advanced too far.

Complete isolation and quarantine of infected pens to prevent spread of
the disease to normal poults is the most important factor in the control pro-
gram. Remedies either in the drinking water or feed should be avoided.

*Keeping the poults warm by increasing the heat in the brooder house, and increased effort to keep them comfortable are essential.* Removal, and destruction by burial or burning, of all dead poults several times daily, and daily dry cleaning of the houses and yards are essential to prevent spread of the infection. Efforts to prevent spread from sick pens to well pens will be much more profitable than time spent in mixing remedies or medicated mashes.

No treatment yet tried in controlled experiments has been effective. Drugs and combinations of drugs that have been tried experimentally include mercuric chloride (1:8,000, and 1:4,000 as a substitute for drinking water) (McNeil and Hinshaw, 1945) ; sodium bicarbonate (baking soda), copper sulfate, nicotine sulfate, iodine, sulfonamides, penicillin, and several arsenical preparations. None gave any promise either for preventing or controlling the disease. A large number of proprietary remedies sold to turkey growers have also been tried under experimental and field conditions, and all have proven equally ineffective. Some of the drugs tried proved toxic for poults when given in dosages recommended.

## REFERENCES

California State Department of Agriculture: 1945. Hexamitiasis in peafowl. Ann. Rep. Petaluma Poultry Path. Lab., Calif. St. Dept. Agr., Monthly Bul. 30:455.

Campbell, J. G.: 1945. An infectious enteritis of young turkeys associated with Cochlosoma sp. Vet. Jour. 101:255.

Doyle, L. P., Cable, R. M., and Moses, H. E.: 1947. A destructive turkey disease. Jour. Am. Vet. Med. Assn. 111:57.

Hinshaw, W. R., and McNeil, E.: 1941. Carriers of *Hexamita meleagridis.* Am. Jour. Vet. Res. 2:453.

―――― and McNeil, E.: 1942. Hexamita sp. from the ring-necked pheasant. Jour. Am. Vet. Med. Assn. 101:503.

――――, McNeil, E., and Kofoid, C. A.: 1938a. The presence and distribution of Hexamita sp. in turkeys in California. Jour. Amer. Vet. Med. Assn. 93:160.

――――, McNeil, E., and Kofoid, C. A.: 1938b. The relationship of Hexamita sp. to an enteritis of turkey poults. Cornell Vet. 28:281.

Jungherr, E., and Gifford, R.: 1944. Three hitherto unreported turkey diseases in Connecticut: erysipelas, hexamitiasis, mycotic encephalomalacia. Cornell Vet. 34:214.

Kimura, G. G.: 1934. *Cochlosoma rostratum* sp. nov. an intestinal flagellate of domestic ducks. Trans. Am. Micr. Soc. 53:102.

McNeil, E., and Hinshaw, W. R.: 1941a. The occurrence of Hexamita (*Octomitus*) *columbae* in pigeons in California. Jour. Parasit. 27:185.

―――― and Hinshaw, W. R.: 1941b. Experimental infection of chicks with *Hexamita meleagridis.* Cornell Vet. 31:345.

―――― and Hinshaw, W. R.: 1942. *Cochlosoma rostratum* from the turkey. Jour. Parasit. 28:349.

―――― and Hinshaw, W. R.: 1945. Effect of mercuric chloride on turkeys and on *Hexamita meleagridis.* Poultry Sci. 24:516.

――――, Hinshaw, W. R., and Kofoid, C. A.: 1941. *Hexamita meleagridis* sp. nov. from the turkey. Am. Jour. Hyg. 34: (Sec. C) 71.

――――, Platt, E. D., and Hinshaw, W. R.: 1939. Hexamita sp. from quail and from chukar partridges. Cornell Vet. 29:330.

Stover, D. E.: 1943. Hexamita sp. from the ring-necked pheasant transmissible to turkeys. Jour. Am. Vet. Med. Assn. 103:37.

## LEUCOCYTOZOON INFECTIONS

Smith (1895) discovered a protozoan parasite in the blood of turkeys, which later was named by Volkmar (1930), *Leucocytozoon smithi*. According to Wenyon (1926), Laveran and Lucet in 1905 found a similar blood parasite in turkeys in France. Volkmar in 1930 reported the parasite in turkeys from Minnesota and North Dakota, and Skidmore (1932) reported an outbreak of disease in turkeys from Nebraska caused by *Leucocytozoon smithi*. Skidmore presented evidence that black flies identified as *Simulium occidentale* (Townsend) were the transmitters of the parasite from turkey to turkey. According to the 1943 and 1945 reports of the Committee on Transmissible Diseases of Poultry for the U. S. Livestock Sanitary Association, Leucocytozoon infection in turkeys has also been diagnosed in the following states: Alabama, California, Georgia, Maryland, Michigan, Missouri, Texas, and Virginia.

Johnson and Underhill (1935, 1937a, 1937b), Johnson, Underhill, Cox, and Threlkeld (1938), Travis, Goodwin, and Gambrell (1939), and West and Starr (1940) have published the results of extensive studies on Leucocytozoon infections and possible carriers. Other published reports on the disease in North America include the following: California, by Hinshaw and McNeil (1943); Texas, by Banks (1943); and Manitoba, Canada, by Savage and Isa (1945).

For a complete description of the parasite and the disease, the reader is referred to Johnson, Underhill, Cox, and Threlkeld (1938) and to Johnson (1942).

**Symptoms.** Poults under twelve weeks are most affected. Loss of appetite, droopiness, and a tendency to sit are common symptoms. Visible symptoms seldom last over 2 or 3 days, after which the birds either die or start to recover. When disturbed they move with difficulty, and in the later stages may fall over, gasp, go into coma, and die.

Recovered birds may suffer no serious after effects, but they may carry the parasite in the blood for months. Some individuals develop a chronic type of the disease. Male birds carrying large numbers of the organism in the blood rarely strut or pay any attention to the females.

Moist tracheal râles are common in chronic cases, and the affected individuals make repeated attempts to clear their throats when excited. Death may result from undue excitement or handling of such birds.

**Autopsy findings.** Slight inflammation of the duodenum is the only consistent gross lesion noted in young birds. Anemia, and various degrees of emaciation may be noted, the flesh is flabby, and the musculature is of a brownish color.

In adult carriers no gross lesions are seen as a rule, but occasionally the liver is icteric, enlarged, and cirrhotic. Johnson and his associates believe

that the respiratory symptoms are due to circulatory obstruction by large numbers of parasites resulting in anemia of some of the vital organs.

**Diagnosis.** Microscopic examination of the tissues of diseased birds reveals large numbers of the parasites (Fig. 40.39). Dried blood smears stained according to Wright's or Giemsa's method make satisfactory specimens for examination. Finding one of the species of Simulium feeding on turkeys is further evidence that a Leucocytozoon is involved.

**Transmission.** Skidmore (1932), Johnson *et al.* (1938), and Underhill (1939) have shown that at least three species of Simulium, *Simulium occidentale, S. nigroparvum,* and *S. slossonae* may transmit leucocytozoa by biting turkeys. These are very small stout-bodied, black flies which live along streams. (See also chapter on Ectoparasites.)

**Prevention and control.** No satisfactory treatment has been reported. Confinement-rearing in houses screened against simuliids until the poults are several weeks old is considered practical by Johnson (1939). It is necessary to make the houses for such method of rearing fly proof by the use of cheesecloth. Screen of 16 mesh to the inch failed to keep the flies from entering houses.

Complete segregation of breeding and brooding operations will do much to prevent transmission from adult carriers to poults. Selling of adult breeders, before the poults that are to be kept for replacements are hatched, is recommended.

Johnson *et al.* (1938) failed to find the parasites in pheasants, ruffed grouse, crows, hawks, and buzzards, but did find them in wild turkeys. Travis, Goodwin, and Gambrell (1939) examined chickens, peafowls, guinea hens, and ducks on infected ranches and found them to be negative for leucocytozoa. They found wild turkeys infected in Missouri, Georgia, and Florida. Therefore, wild turkeys may be a source of infection, and their control must be considered in a prevention program. The same precautions recommended for hexamitiasis will also help in preventing this disease.

## REFERENCES

Banks, W. C.: 1943. *Leucocytozoon smithi* infection and other diseases of turkey poults in central Texas. Jour. Am. Vet. Med. Assn. 102:467.

Hinshaw, W. R., and McNeil, E.: 1943. Leucocytozoon sp. from turkeys in California. Poultry Sci. 22:268.

Johnson, E. P.: 1939. A method of raising turkeys in confinement to prevent parasitic diseases. Va. Agr. Exper. Sta., Bul. 323.

——: 1942. Further observations on a blood protozoon of turkeys transmitted by *Simulium nigroparvum* (Twinn). Am. Jour. Vet. Res. 3:214.

—— and Underhill, G. W.: 1935. The recent turkey disease. Southern Planter 96:28.

—— and Underhill, G. W.: 1937a. A blood disease of turkeys. Southern Planter 98:34 and 39.

—— and Underhill, G. W.: 1937b. A blood disease of turkeys. Southern Planter 98:32.

——, Underhill, G. W., Cox, J. A., and Threlkeld, W. L.: 1938. A blood protozoon of turkeys transmitted by *Simulium nigroparvum* (Twinn). Am. Jour. Hyg. 27:649.

Savage, A., and Isa, J. M.: 1945. An outbreak of Leucocytozoon disease in turkeys. Cornell Vet. 35:270.

Skidmore, L. V.: 1932. *Leucocytozoon smithi* infection in turkeys and its transmission by *Simulium occidentale* (Townsend). Zentralbl. f. Bakt. I. Orig. 125:329.

Smith, T.: 1895. Infectious diseases among poultry. U.S.D.A., Bur. An. Ind., Bul. 8:7.

Travis, B. V., Goodwin, Jr., M. H., and Gambrell, E.: 1939. Preliminary note on the occurrence of *Leucocytozoon smithi* Laveran and Lucet (1905) in turkeys in southeastern United States. Jour. Parasit. 25:278.

Underhill, G. W.: 1939. Two Simuliids found feeding on turkeys in Virginia. Jour. Econ. Ent. 32:765.

Volkmar, F.: 1930. Observations on *Leucocytozoon smithi;* with notes on leucocytozoa in other poultry. Jour. Parasit. 16:24.

Wenyon, C. M.: 1926. Protozoology. William Wood and Co., New York.

West, J. L., and Starr, L. E.: 1940. Further observations on a blood protozoan infection in turkeys. Vet. Med. 35:649.

## TRICHOMONIASIS OF THE UPPER DIGESTIVE TRACT

### (Necrotic Ulceration of the Crop, Jungherr's Disease)

Volkmar (1930) described a species of Trichomonas which he calls *Trichomonas diversa,* associated with necrotic ulceration of the crop (Jungherr's disease). Hawn (1937) has shown that Trichomonas is the etiological agent in this disease. As far as is known this species has not been found in turkeys posterior to the proventriculus. Gierke (1933) described necrotic ulceration of the upper digestive tract of chickens associated with a heavy infection of trichomonads similar to those described in turkeys by Volkmar and by Hawn.

Jungherr (1927) was the first to describe this disease. At that time he suggested that a fungus was the probable cause. Three years later Volkmar (1930) reported that *Trichomonas diversa* is a constant inhabitant of the crops of turkeys suffering from this disease, and as mentioned above, Hawn has shown these parasites to be the etiological agent. Stabler (1938a, 1938b) has shown the similarity of this trichomonad to the one in pigeons which he points out should be known as *T. gallinae* instead of *T. columbae.* He suggests that the species in turkeys be called by this name instead of *T. diversa.* The name *T. gallinae* must not be confused however with *T. gallinarum,* a separate species inhabiting only the lower intestine of fowls. Levine, Boley,

---

Fig. 40.39. Photomicrographs of stained turkey blood containing various stages of Leucocytozoon from turkeys. ×750. Giemsa's stain.

1—a microgametocyte with only one lateral bar present. Note light color of parasite.

2—a macrogametocyte with only one lateral bar present. Note dark color of parasite.

3—a macrogametocyte with one bar on one side and two bars on opposite side.

4—the microgametocyte shown at *a,* and the macrogametocyte at *b,* each with bilateral bars, are the most common forms found. At *c* may be seen a macrogametocyte that has become round to form a macrogamete. Note one bar is still attached ventrally.

5—an early microgametocyte with distinct difference between density of central body, or what might be the parasite proper, and the surrounding cytoplasm connecting the two lateral bars or possible host-cell nucleus. At the right and ventrally is a typical granulocyte with eosinophilic rod granules. Note comparative size.

6—the earliest macrogametocyte found. Here, again, there is well-marked distinction between central body and surrounding cytoplasm including the bilateral bars. (Johnson *et al.,* Am. Jour. Hyg.)
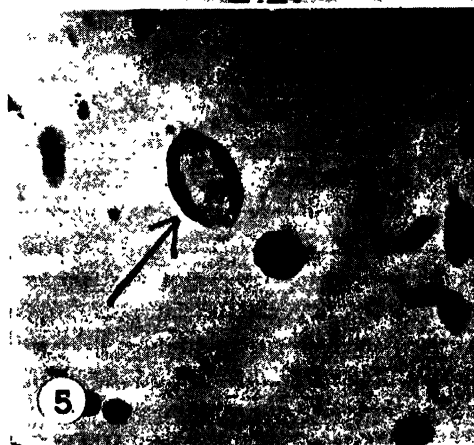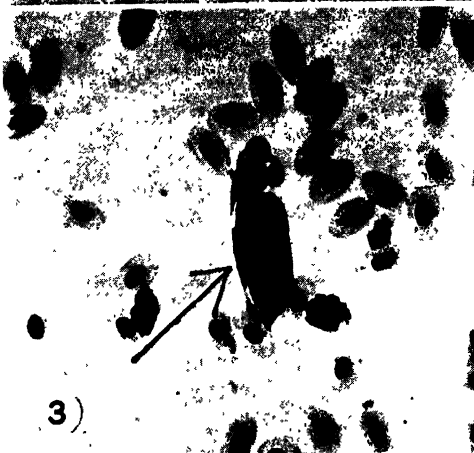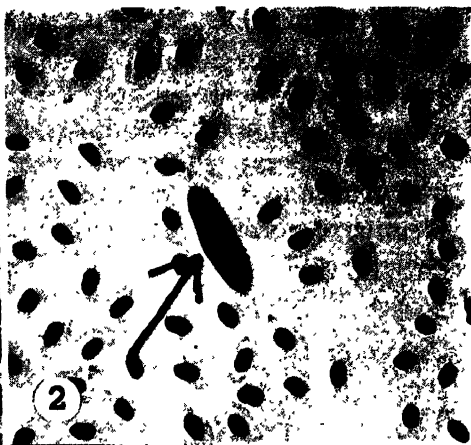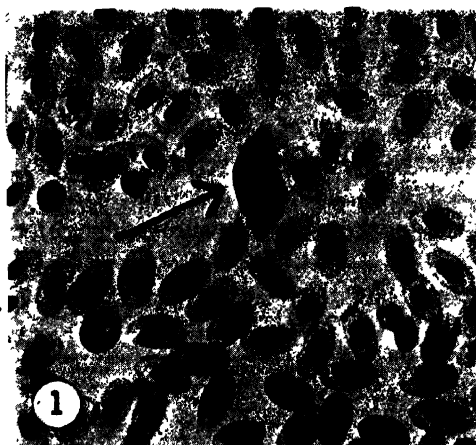
and Hester (1941) summarized the literature on this disease in birds and report further evidence that the species found in turkeys is the same as that found in chickens and pigeons. They were able to produce the typical disease in turkeys, chickens, bobwhite quail, canaries, and English sparrows with a species isolated from chickens.

**Epidemiology.** Most of the cases studied by Hinshaw have been in turkeys from sixteen to thirty weeks of age reared on range land. The following description of one outbreak in California is typical of the environment in which most of the cases are found:

The turkeys involved in this outbreak had been reared on the home



Fig. 40.40. Posture typical in trichomoniasis of the crop. Note especially the sunken appearance of the crop area. (Hinshaw, Univ. of Calif.)

ranch under semiconfinement methods until the middle of September, when they were driven daily to a cutover rice field about ½ mile from the ranch. Each day the birds were allowed to feed for 2 or 3 hours on the shattered rice left by the harvester. After this procedure had been continued for about a month, the flock was permanently moved to the rice field and allowed to range at will. They were fed a mash supplement that was left near the roosts located on a dry area in a cutover barley field adjacent to the rice. Water was hauled from the home ranch, but the birds had access to the sluggish, algae-contaminated water in an irrigation ditch, which they had to cross in order to reach the rice from the roosting and mash-feeding areas. Seepage from the ditch had caused a large, muddy stagnant pool to form near the edge of the rice field. The turkeys drank a great deal of this water and picked up the rice in the mud at the edge of the pool. Several similar pools were found in other parts of the field. The disease started within 10 days after the birds were permanently located on the rice field. Pigeons, abundant in the area, may have been the transmitting agent.

**Symptoms.** The symptoms are similar to those seen in many other diseases. Darkened heads with sunken sinuses and a generally haggard appear-

ance are characteristic. The chest always has a depressed appearance, with the crop empty and drawn in towards the body. This typical attitude is seen in Figure 40.40. Lack of appetite, drooling from the mouth, roughened un-kempt feathers, and a normal or slightly subnormal temperature are also observed. Diarrhea does not, as a rule, accompany the disease. A foul odor is always present. The course of the disease varies, but as a rule, it is pro-longed, and the birds become emaciated before death.

**Autopsy findings.** Chronic ulceration of the crop is the most common autopsy finding. The lower esophagus and, less often, the proventriculus and upper esophagus may also be involved. The lower digestive tract and the
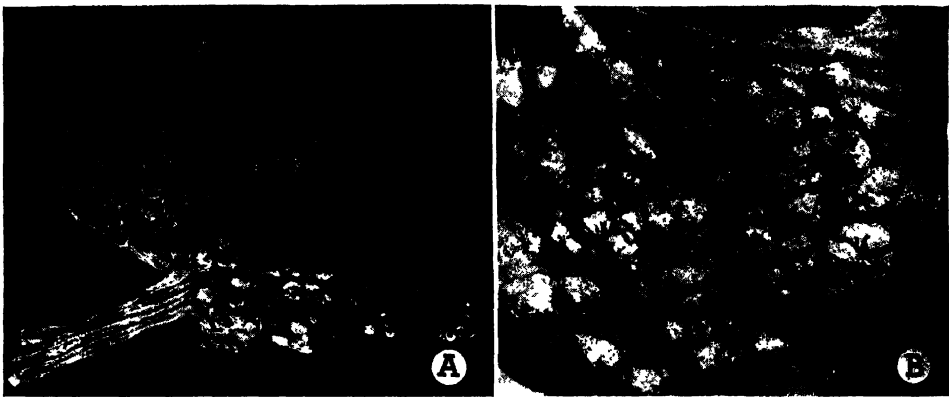


FIG. 40.41. A—necrotic ulceration of the esophagus and crop seen in trichomoniasis. B—close-up of typical pyramid-like necrotic ulcers characteristic of trichomoniasis of the upper digestive tract. (Hinshaw, Univ. of Calif.)

other organs are, as a rule, normal. Aspergillosis of the lungs may be second-ary to the necrotic ulceration of the upper digestive tract.

The lesions involve the glandular tissue and vary in size from a few to 15 millimeters in diameter at the base (Figs. 40.41 A and B, and 40.42). They taper to a point in concentric rings of piled-up necrotic tissue to as much as 5 millimeters above the surface. They may extend into the tissue 3 or 4 millimeters. The surface protruding into the lumen of the organ is rough, irregular, and surrounded at the base by a circular hemorrhagic ring. The lesions in the esophagus are usually smaller than those in the crop but are similar in shape and structure. When the proventriculus is involved, the esophageal portion is most affected. The lesions in the proventriculus are, as a rule, coalesced and may appear as a solid ring of necrotic material causing a marked thickening of the tissues and resulting in partial to complete occlu-sion of the lumen. In many such cases, impactions of the lower esophagus have been noted.

**Prevention and control.** Since the disease is directly associated with in-

sanitary surroundings, sanitation is of primary importance. In addition, adequate supplements must be fed to all turkeys ranged on cutover grain fields.

The disease can be prevented by keeping turkeys away from known infected areas, and by avoiding as range lands poorly drained fields where stagnant pools of water exist or where birds have access to stagnant, sluggish irrigation ditches. Old strawstacks that have become decomposed should be avoided, as should areas of grain fields that have been beaten down and covered with water for any length of time. Contact with pigeons should also be avoided.



Fig. 40.42. Necrotic ulceration of the proventriculus often seen in trichomoniasis of this organ. (Hinshaw, Univ. of Calif.)

The first requisite for control is sanitation. As soon as the disease is observed, the flock should be moved to a dry, clean area and given plenty of pure, fresh water to drink. Sick birds should be kept separate and cared for by a person who has no contact with the healthy birds. Removing the causal agent and giving the birds good care is more essential than treatment with drugs. The use of a 1:2,000 solution of copper sulfate in place of all drinking water is the only treatment tried that has met with any success. If copper sulfate solution is used, it should be kept before the birds for 2 or 3 days and then repeated after a few days, if improvement has not been noted. All other sources of water must be removed during such treatment.

## REFERENCES

Gierke, A. G.: 1933. Trichomoniasis of the upper digestive tract of chickens. Calif. St. Dept. Agr., Mo. Bul. 22:205.

Hawn, M. C.: 1937. Trichomoniasis of turkeys. Jour. Infect. Dis. 61:184.

Jungherr, E.: 1927. Two interesting turkey diseases. Jour. Am. Vet. Med. Assn. 71:636.

Levine, N. D., Boley, L. E., and Hester, H. R.: 1941. Experimental transmission of *Trichomonas gallinae* from the chicken to other birds. Am. Jour. Hyg. 33: (Sec. C) 23.

Stabler, R. M.: 1938a. The similarity between the flagellate of turkey trichomoniasis and *T. columbae* in the pigeon. Jour. Am. Vet. Med. Assn. 93:33.

———: 1938b. *Trichomonas gallinae* (Rivolta, 1878) the correct name for the flagellate in the mouth, crop, and liver of the pigeon. Jour. Parasit. 24:553.

Volkmar, F.: 1930. *Trichomonas diversa* n. sp. and its association with a disease of turkeys. Jour. Parasit. 17:85.

## MISCELLANEOUS PROTOZOAN DISEASES

Protozoan infections that have been reported one or more times, and which may in the future prove to be significant causes of mortality are briefly described below.

A Cochlosoma, first described by Kotlán (1923) from ducks in Europe, was named by him *C. anatis*. Kimura (1934) described a species from American ducks which he called *C. rostratum*. Travis (1938), in a synopsis of the genus, considers these species synonymous. McNeil and Hinshaw (1942) described a species in turkeys which was apparently the same species as described by Kotlán and Kimura. Campbell (1945) found Cochlosoma in turkey poults in Scotland. The true pathogenic significance of this flagellate is not known, although both Kotlán and Campbell suggest that it may be the cause of enteritis. Hinshaw has always found it associated with Hexamita or Salmonella.

Haemaproteus has been found by Morehouse (1945) in turkeys in Texas. Banks (1943) also suggested its presence in turkeys in Texas. This genus is usually carried by flies of the genus Pseudolynchia. Morehouse does not mention the insect vector for the species found in turkeys. Rivero (1947) has reported that the cone-nosed bug, Triatoma, is able to transmit *Haemoproteus columbae*.

Herman (1941) described *Plasmodium durae* as the cause of bird malaria in a turkey in Kenya Colony, British East Africa.

McNeil and Hinshaw (1944) found an intraerythrocytic parasite in turkey poults. The organism probably belongs in the Babesiidae.

For a more detailed discussion of these diseases, the reader is referred to the chapter on Protozoa.

## REFERENCES

Banks, W. C.: 1943. *Leucocytozoon smithi* infection and other diseases of turkey poults in central Texas. Jour. Am. Vet. Med. Assn. 102:467.

Campbell, J. G.: 1945. An infectious enteritis of young turkeys associated with Cochlosoma sp. Vet. Jour. 101:255.

Herman, C.: 1941. *Plasmodium durae*, a new species of malaria parasite from the common turkey. Am. Jour. Hyg. 34:22.

Kimura, G. G.: 1934. *Cochlosoma rostratum* sp. nov. an intestinal flagellate of domesticated ducks. Trans. Am. Micr. Soc. 53:102.

Kotlán, A.: 1923. Zur Kenntnis der Darmflagellaten aus der Hausente und anderen Wasservögeln. Zentralbl. Bakt. I. Abt. Orig. 90:24.

McNeil, E., and Hinshaw, W. R.: 1942. *Cochlosoma rostratum* from the turkey. Jour. Parasit. 28:349.

——— and Hinshaw, W. R.: 1944. A blood parasite of the turkey. Jour. Parasit. (Supplement) 30:9.

Morehouse, N. F.: 1945. The occurrence of Haemoproteus sp. in the domesticated turkey. Trans. Am. Micr. Soc. 64:109.

Rivero, M. D.: 1947. La infección experimental por el *Haemoproteus columbae* Celli y Sanfelice. Rev. Med. Mexicana 26:197.

Travis, B. V.: 1938. A synopsis of the flagellate genus Cochlosoma Kotlán, with the description of the two new species. Jour. Parasit. 24:343.

## MISCELLANEOUS DISEASES

This section includes diseases and conditions which cause considerable financial loss in certain flocks but which are more or less sporadic in nature.

### ABSCESS OF THE FOOT PADS
#### (Bumblefoot)

Turkeys sometimes suffer from abscesses of the foot pads (Fig. 40.43).



FIG. 40.43. Abscesses of the foot pads (bumblefoot). (Hinshaw, Univ. of Calif.)

These may resemble corns and are similar to a condition, commonly called bumblefoot, in chickens. The cause is not known, but the abscess probably starts from an infection following an injury to the foot pad. Some of the cases observed have resembled foot rot as seen in other animals. In these instances the affected turkeys had been in yards that were in constant use and which were covered with several months' collection of feces; cases usually appeared after the fall rains when the yards became very muddy. No doubt many cases of abscesses of the foot pads are also identical with staphylococcal arthritis.

Putting the birds in clean dry quarters and treating the diseased pad will cure many cases. If pus is present, it should be removed, and the area cleaned and treated with an antiseptic healing ointment or tincture of iodine. Ammoniated mercurial ointment is an example of a satisfactory ointment.

Rotating the runs and removing the birds to a clean, well-drained yard just before the breeding season are recommended as preventive measures.

## ASCITES

Although not a hatchery problem, ascites caused by excess of sodium compounds including common salt (Scrivner, 1946), (Doll, Hull, and Insko, 1946), and by exposure to certain disinfectant fumes (Bullis and Van Roekel, 1944), are sometimes confused with omphalitis due to infection described by Brandly (1932). Ascites (dropsy, watery belly, etc.) due to chemical poisoning is seen in young poults or chicks from 2 or 3 days to two weeks of age. Excess of salt in the ration and fumes from certain types of disinfectants used for spraying brooder floors are common causes. Excessive salt is usually accidental and may be due to improper mixes, or poor screening which allows lumps to get into the mix. The addition of salt to mashes which already contain it because of salted protein concentrates has also been responsible for a few outbreaks. Hatcherymen should warn customers regarding excessive salt in mashes and of the dangers of putting chicks into brooders too soon after spraying the floors with volatile disinfectants.

## REFERENCES

Brandly, C. A.: 1932. An acute infectious omphalitis of baby chicks. Poultry Sci. 11:279.

Bullis, K. L., and Van Roekel, H.: 1944. Uncommon pathological conditions in chickens and turkeys. Cornell Vet. 34:312.

Doll, E. R., Hull, F. E., and Insko, Jr., W. M.: 1946. Toxicity of sodium chloride for baby chicks. Vet. Med. 41:361.

Scrivner, L. H.: 1946. Experimental edema and ascites in poults. Jour. Am. Vet. Med. Assn. 108:27.

## BLUEBACKS AND CANNIBALISM

Blueback, as the name indicates, is a condition in which the backs of the affected turkeys are discolored blue or black. According to Billings (1940), it is caused by an injury to the quills of the feathers at the point of entrance into the skin which allows the pigment to escape and tattoo the surrounding skin (Fig. 40.44). Feather picking is the immediate cause, according to him. Exposure to sunlight after picking is necessary to produce the pigmented condition. Some of the other causes are overcrowding in the brooder, keeping the poults too long on the sun porch, and lack of sufficient fiber in the ration. After picking becomes a habit, the vice is difficult to control, and the financial loss due to lowering of the market grade of the carcass may be considerable. Another form of cannibalism which often results in evisceration may also be started by feather picking.

Prevention and control consist in correcting the vice. Overcrowding in the brooder should be avoided. Moving poults to the range as soon as picking starts or reducing the numbers in a house are suggested means of control. The feeding of whole oats is recommended by some as another means of prevention.

Mechanical devices are commonly used for the prevention of these vices.

Two types are used. The first, inserted in the beak, is patterned after the hog nose ring, in use by swine raisers for prevention of "rooting." These are inserted in one side of the lower mandible (lower beak). A similar type sold by one manufacturer is pinned in the upper beak. The promoters of these nose guard types claim that turkeys so fitted cannot pick feathers.

Trimming the edges of the beaks will temporarily prevent picking. An electrically heated cauterizing knife has been developed for removing a portion of the upper beak of birds to prevent feather picking and cannibalism. This instrument can be recommended for the prevention of these vices in turkeys.

Several ointments are on the market for use on injured birds, principally for prevention of feather picking. These usually consist of a vaseline base, some bitter drug such as aloes, and a red coloring like carmine. Ewing (1940) suggests an ointment made by mixing 4 ounces of vaseline, $\frac{1}{4}$ ounce of carmine, and $\frac{1}{2}$ ounce of aloes. Roofing tar is also used by many growers.

### REFERENCES

Billings, W. A.: 1940. Common diseases of turkeys. Minn. Ext. Bul. 214.
Ewing, W. R.: 1940. Handbook of Poultry Nutrition. W. R. Ewing, Upper Montclair, N. J. (Ref. p. 205.)

### ENTERITIS (NONSPECIFIC)
#### (Inflammation of the Intestines)

Every year numerous immature turkeys die of enteritis from unknown causes. Further research may prove some of these outbreaks to be infectious in nature. At present, however, they must be handled as nonspecific and can probably be attributed to a number of causes.

Stampeding, failure of brooder heaters, sudden changes in the weather, piling in the brooder houses, heat prostrations, sudden changes of feeding methods, and probably, in many cases, faulty feeding methods over a period of several weeks, are examples of obscure causes of mortality, with enteritis as the principal pathological manifestation. These various factors may also pave the way for secondary invasion by microorganisms normally of low virulence, which, under such conditions, may cause heavy mortality.

An example of losses starting from an obscure cause that may easily be overlooked follows: Three lots of turkeys about twelve weeks of age were in similar yards where it had been necessary to use an undesirable watering system until a modern drip system was installed. This new system was installed in all three yards at the same time, and the old system removed. Within 48 hours two of the three lots of turkeys became ill, while the third remained normal. On the third day it was discovered that the two groups of sick birds were not drinking the water because of an apparent fear of the new equipment. When the old equipment was replaced in these pens, the birds

fought to get at the water and drank three or four times as much as normal for the day. Only 1 per cent of the birds died before and within 24 hours after the discovery of the cause, but there was a distinct difference between the two affected lots and the third lot for nearly a month. A difficulty usually experienced in making a diagnosis for such outbreaks is the lack of sufficient history.

Losses are commonly experienced by turkey growers following the transfer of poults from the starting brooders to the "cooling" brooders. A common practice among some growers is to rear poults in small units such as the so-called sunshine type or in battery types for three to four weeks, and transfer from these to regular brooder houses with yards or wire porches. Losses following such transfers are usually due to failure to use the same type of feeders and waterers as in the starting brooders. Use of similar equipment and careful watching of the poults to see that they start eating and drinking properly after such moves will avoid losses.

**Symptoms.** The principal symptoms seen in enteritis are loss of appetite, a tendency to separate from the well birds, diarrhea, and a general haggard appearance. Temperatures are usually normal or subnormal. The birds may sit



FIG. 40.44. "Blueback" of turkeys. (Ralston Purina Co.)

in a listless manner with their heads hung or turned up over their backs. On open ranges, where the majority of the flock is affected, difficulty is often experienced in keeping the birds under control; the turkeys appear nervous and may wander for hours, often straying ½ mile or more from the main camp. During the course of the disease, often a period of several weeks, a marked loss of flesh may occur. The mortality is not, as a rule, high for a single day; but over a period of three or four weeks, 25 per cent or more of the birds may die. The greatest loss, however, results from failure of the birds to recover completely and to make proper gains.

**Autopsy findings.** Emaciation and enteritis, varying from a catarrhal to a more advanced inflammatory type, are the principal autopsy findings. The head has a drawn appearance, with the eyes and sinuses sunken. The heart

is usually flabby. The blood, in many instances, fails to clot for several hours after death; it is usually very dark in appearance. The liver often appears congested, and dark venous blood oozes from cuts made on its surface. In many respects the symptoms and autopsy findings resemble those of acute poisoning.

**Prevention, control, and treatment.** It is extremely difficult to give methods of prevention, control, and treatment for enteritis of an unknown cause. Sound, rational turkey husbandry is probably the best preventive. An adequate diet and an ample supply of pure, fresh water are important.

Avoiding the possible causes of enteritis is essential. A few have already been mentioned, and others will suggest themselves. Any abnormality that will cause the bird to lose its appetite or develop an intestinal disturbance, even for a few days, may cause heavy losses for several weeks.

The successful feeder of any class of livestock realizes the need for constant attention to the flock or herd to detect the first symptoms of failure to make proper gains. Sudden changes of feed should be avoided, but if the flock is definitely not doing well on a particular diet, the reason should be sought. If the feed is responsible, a gradual change to another method should be made. If the original method is resumed after the birds have recovered, the shift should also be gradual.

A common fault of turkey growers is to supplement an already adequate commercial growing mash with milk, fish meal, or meat scraps, which increases a protein level already near the maximum tolerance for turkeys.

If cheaper sources of high-protein feeds than are in a commercial product are available, a properly balanced feed should be obtained to mix with the available supply. An equally common mistake and one that may cause very severe losses is to remove all mash supplement when turkeys are moved to barley, wheat, or rice ranges. The first month on a new range is probably the most important one, and no doubt more losses are experienced from failure of the birds to become properly adapted to the new environment than for any other reason. As the average grain field does not contain an adequate supply of all the necessary food elements for proper growth, a supplement is necessary. The most important elements likely to be deficient on a cutover grain field are greens for vitamins A and B and protein concentrate. The amounts of each that are needed will depend on the amount of green grass and insect life available. Each range constitutes an individual problem and must be studied carefully; a suitable concentrate must be furnished the birds.

Turkeys that are to be taken off a full-feed ration and transferred to a grain field should be fed some of the same type of grain as that grown in the field for a week or two before being moved to the range. This procedure accustoms them to the new grain and will prevent a setback and possible heavy

losses. In addition, for a few days after they are moved to the range, the birds should have some of the mash previously used.

### ENTERITIS (HEMORRHAGIC)

Pomeroy and Fenstermacher (1937) describe a hemorrhagic enteritis in turkeys ranging from seven to twelve weeks of age. The disease appeared in Minnesota during the summer months and caused a mortality of about 10 per cent. The flocks involved in this outbreak had been reared on wire porches for six to eight weeks and were then transferred to field ranges. The losses occurred from 10 to 14 days after the poults were put on range. The ranges were very poor that year, greens were not available in sufficient quantities, and there was a distinct lack of sunshade and shelter. *Escherichia coli* and an unidentified Gram-positive rod were the only bacteriological findings. Neither of these was proved to be the causal factor.

A condition similar to that reported by Pomeroy and Fenstermacher, has been observed in a few instances by Hinshaw. In these outbreaks the losses have always occurred a few days after transferring the poults to ranges or to yards adjoining the brooder. In two such outbreaks the range contained a young succulent growth of alfalfa, and in a third the yard was overgrown with weeds, and grasses including some sweet clover. Losses stopped in all three instances when the poults were returned to the brooders, and put back on a dry mash ration. Later they were put back on the range for short intervals at first and finally for the full period and without further losses. The causal factors were not determined.

These findings constitute additional reasons for use of great precaution when moving poults from one environment to another. (See prevention, control, and treatment under Enteritis—Nonspecific.)

### REFERENCE

Pomeroy, B. S., and Fenstermacher, R.: 1937. Hemorrhagic enteritis in turkeys. Poultry Sci. 16:378.

### HEAT PROSTRATION
(Heat Stroke)

Heat prostration is usually associated with high humidity accompanying high temperatures or with very low humidity on excessively hot days. Losses from this cause most often occur in young turkeys that have recently been moved from the brooder house to a range having inadequate shade. Stiles (1943) reported a case of heat exhaustion in a flock of three-week-old turkey poults which were abruptly transferred from cool battery brooders to quarters where the heat inside the building and on the sun porches was unbearably hot.

The symptoms are labored breathing, weakness, excessive thirst, and high temperature, followed by complete prostration. Losses can be prevented by furnishing ample shade facilities, especially for the poults just transferred from the brooder house to an open yard or range. If a house is available on the range, the young poults may well be sheltered in it during the hottest part of the day, but with all the windows open for ample circulation of air. Plenty of water should be available. As soon as the poults become accustomed to the new quarters, they will stay inside during the excessive heat; water and feed should be left both inside and outside the house for the first few weeks. Out of doors, trees make the best shade; but an abundance of cheap artificial shade can be made from old lumber and posts. Thatched roofs may be used advantageously if material for covering the shelter can be secured. Pure, fresh water must be available at all times. It should be kept in a shady place, in enough containers so that the birds will have no difficulty in getting to it. If, in spite of all precautions, turkeys are overcome by the heat, they should be put in a shady, well-protected place and sprayed with cold water. Used in time, this procedure will save a large number. Filling the crop with cold water by means of a rubber tubing and a funnel is also advisable. Dipping



FIG. 40.45. Turkey hen with a severe laceration caused by a male during the mating process. (Hinshaw, Univ. of Calif.)



FIG. 40.46. A—a method of trimming the toenails of a male turkey to prevent injuries during the mating season. B—feet of a male turkey after trimming the toenails as illustrated in A. (Hinshaw, Univ. of Calif.)

the birds in cold water may be effective, but care must be taken to prevent drowning. As they may remain weak for several days, they should be kept in the shade with food and water easily accessible.

<div style="text-align:center">REFERENCE</div>

Stiles, G. W.: 1943. Heat exhaustion in young turkeys. Poultry Sci. 22:242.

<div style="text-align:center">INJURIES</div>

**Injury to the female by the male.** Severe losses occur in many breeding flocks because of the females' backs being badly torn by the male during the mating process (Fig. 40.45).



Badly torn females seldom recover sufficiently to produce fertile eggs during the remainder of the season; and if the wound does heal, the area is tender and easily torn when the bird is trodden again.

Some males are much more vigorous and rough in the mating than others, and many of the losses can be traced to one or two individuals in a flock. These males should immediately be replaced by reserves. One method of prevention is the removal of the toenails from the males (Fig. 40.46 A and B). This should be done about a week before the males are put into the breeding pens. A convenient

FIG. 40.47. A type of "apron" or "saddle" in common use for prevention of injuries to females during the mating season. See Figure 40.48. (Hinshaw, Univ. of Calif.)

instrument for removing the toenails is a pair of pruning shears of the roll-cut type shown in Figure 40.46 A. An electric soldering iron or some other form of a searing iron can be used for searing the cut surface to stop hemorrhage after the operation. It is a good plan to smooth off the edges of the cut surface with a file or sandpaper just before the male is placed in the breeding pen.

Another method of preventing breeding females from being torn is to fit a canvas jacket over the back (Figs. 40.47 and 40.48). These jackets, which can be purchased at a reasonable price, are recommended for general use. Care should be taken to purchase saddles which fit correctly in order to prevent strangulation, or injury to the body or wings.

If an injured hen is discovered immediately, the torn edges of skin should

be sutured with a heavy thread dipped in iodine. The birds should then be placed in a pen where there are no males and left for about two weeks. An antiseptic dusting powder such as boric acid, sodium perborate, or one containing a sulfonamide will induce healing and prevent attacks by flies. As soon as the wound begins to heal normally, the hen can be fitted with the canvas jacket described above, and can be returned to the breeding pen. She should be carefully watched, however. and if again injured by the males should be returned to the isolation pen.



FIG. 40.48. Turkey hen with the "saddle" shown in Figure 40.47 in place. (Hinshaw, Univ. of Calif.)

Wounds that are not discovered for several days seldom respond well to treatment. They should be carefully cleansed and washed with a mild antiseptic solution and treated with an antiseptic dusting powder. It may be necessary to make an incision in the skin below the wound for drainage and to trim necrotic edges of the skin around the wound. The birds should be kept out of the breeding pens for two or three weeks and treated daily. They should be jacketed before being returned. Where several males are in one pen, the transfer of a male to a pen of injured females may be a better procedure than putting the injured hens back in the regular pen. The time required for complete recovery depends on the extent of the injury and the efficiency of the treatment. Whether or not treatment is worth while depends on the value of the individual and the time available.

**Injuries from fighting.** As males are more likely to be injured from fighting than are females, often a valuable male should be separated from its pen mates if it is not able to defend itself successfully. A male which has been away from the flock for any length of time or



FIG. 40.49. Posture of turkey suffering from a slight dislocation of one vertebra of the neck. The bird could not raise its head and the muscles of the area were severely swollen. (Hinshaw, Univ. of Calif.)

which has just been purchased must be protected when placed with other males, because they will invariably fight it.

Minor injuries seldom require treatment and will heal readily if the bird is unmolested. Severe lacerations about the head usually respond to iodine or an antiseptic dusting powder. If flies are troublesome, carbolized vaseline may be used to cover the wounded areas.

**Miscellaneous injuries.** Injuries from being caught in fences, from flying into objects during stampedes, from rough handling, and from many other causes are cared for in much the same manner as injury by a male.

Wickware (1945) reported that grasshoppers may cause death of turkeys by mechanically injuring the walls of the crop and intestine. In some of these cases the walls of these organs were punctured by grasshopper legs. He suggests that such losses can be prevented by feeding plenty of mash to turkeys that have access to ranges where grasshoppers are abundant. Dickinson and Clark (1946) and Bullis and Van Roekel (1944) have reported brooder stove residue burns on the heads of turkey poults that were brooded under stoves heated with gas briquets or kerosene. The injuries in these cases are similar and range from mild burns to dry gangrene. Dickinson and Clark state that the use of tight-fitting stove pipe joints will prevent seepage of the oily residue responsible for gas briquet type burns. Attention paid to kerosene or similar type burners to prevent leakage of oil will stop losses from such causes.

A type of injury seen a few times is shown in Figure 40.49; the bird is usually found with its head hanging downward and forward and is unable to change this position. The neck muscles are much swollen and are hot to the touch. Often one will find a tuft of feathers pulled from the side of the neck and evidence of a bruise. Dislocation of a vertebra or fracture of a vertebral process has been found to be the cause in most cases. In at least two, the injury resulted from entanglement in a wire fence during atempts to reach feed. Correction of the dislocation by massage and tension gave relief and complete recovery in about two weeks. Other cases have taken from three to six weeks to recover but have shown no detrimental after-effects. If such an injury is found in a flock, the cause should be determined. Any dislocation found should be corrected. Until recovered the bird should be isolated but placed near water and feed containers.

## REFERENCES

Bullis, K. L., and Van Roekel, H.: 1944. Uncommon pathological conditions in chickens and turkeys. Cornell Vet. 34:312.

Dickinson, E. M., and Clark, W. G.: 1946. Brooder-stove-residue burns on turkey poults. Cornell Vet. 36:314.

Wickware, A. B.: 1945. Grasshoppers, a potential danger to turkeys. Canad. Jour. Comp. Med. 9:80.

## OMPHALITIS

Omphalitis, or navel infection, is characterized by failure of the navel opening (umbilicus) to close properly, with resultant infection of the internal organs. The disease can often be traced to faulty incubation or to hatchery insanitation. In most instances the poults are weak when removed from the incubator, and losses may start before time for shipment from the hatchery.

The symptoms are general weakness, lack of body tone, and a tendency to huddle. In the brooder the poults appear cold and stay under the hover. When handled they feel flabby, the abdomen is enlarged, and they do not have the firmness of a normal poult. The navel opening, which usually is completely healed within 72 hours, is inflamed, moist, and fails to close for several days. Often a definite scab forms over the opening. The course is rapid, death often occurring within a day after symptoms are noted; the mortality is high, often reaching 50 per cent of the brood.

On autopsy, edema of the muscles of the abdomen and breast, an unabsorbed yolk, and peritonitis are the principal observations. The contents of the retained yolk are usually more liquid than normal, and rupture of the yolk sac is common.

The disease is probably a result of a mixed infection, of hatchery origin. In the outbreaks reported to the writer, thorough cleaning and disinfection of the hatchery rooms and incubators have prevented further losses. The formaldehyde fumigation method, outlined under the section "Disinfectants" (p. 100) will eliminate the disease from the hatchery. Bittenbender (1940) suggests that twice the amounts of formalin and potassium permanganate be used to fumigate incubators known to be spreading omphalitis. *This strength should be used between hatches.* Incubator rooms and all hatchery equipment as well as the incubators should be fumigated. (See section on Formaldehyde Fumigation, page 104.)

No remedy or adequate method of controlling the disease in the brooder has been found. Keeping the poults comfortable and applying hygienic measures will help reduce the mortality to a minimum.

### REFERENCE

Bittenbender, H. A.: 1940. Incubator fumigation. New England Poultryman 31 (1) :8–9.

## PENDULOUS CROP
### (Water Crop, Dropcrop, Baggy Crop)

Serious losses from pendulous crop (Fig. 40.50 A) in some flocks are, according to recent investigations at this station (Hinshaw and Asmundson, 1936; and Asmundson and Hinshaw, 1938), the result of a hereditary predisposition toward the condition. Turkeys with the inherent weakness develop pendulous crops after the increased liquid intake that follows the first

wave of excessively hot weather. The crop, once expanded, seldom returns. to normal size, especially if the hot, dry weather continues. It may contract for a few days, if the weather becomes cool, and then expand again during the next hot spell. Although a few birds recover, the majority continue to have pendulous crops. In this condition the crop does not empty normally; stagnant, sour liquid contents are retained in the bulbous portion. As time goes on, the mucous membrane thickens and may become ulcerated (Fig. 40.50 B).



FIG. 40.50. A—an 8-month-old female turkey with a pendulous crop of about 5 months' duration. B—section of a pendulous crop showing thickening and ulceration of the mucous membrane. (Hinshaw, Univ. of Calif.)

Although the appetite is not greatly affected, digestion is hindered. The feed and water remaining in the crop may increase until the crop and its contents equal one-fourth of the total live weight of the bird. The bird may continue to grow, but will remain unthrifty and may become emaciated.

Pendulous crops caused by an inherent weakness must be distinguished from similar conditions that sporadically result from impactions, mycosis, trichomoniasis, and other crop infections.

**Course, mortality, and causes of death.** The course of the disease is chronic; as mentioned above, very few birds recover even with treatment. Some live for as long as two years, but the mortality of the affected birds in a flock may exceed 50 per cent.

The causes of death are (1) rupture of the crop by the bird's toes in its attempt to walk or run, (2) mechanical pneumonia from the seepage of crop contents into the bronchi during mechanical efforts to drain the crop or as a result of a back-flow when the bird lowers its head, and (3) starvation due to insufficient intake of food or to improper digestion.

Necrotic ulcers, varying in nature according to the type of the contents

and severity of the case, frequently occur. Scraping the necrotic membrane from the surface leaves a denuded, bleeding area. This type of necrosis is distinguished from that seen in trichomoniasis by the tendency of the latter to form individual pyramidal ulcers as compared with the diffuse, spreading nature of the former. Demonstration of trichomonads furnishes a further means of differentiation. In a few cases, lesions typical of moniliasis (thrush) have also been observed. In these cases fungi are readily demonstrated. The contents of the crops have varied from a watery, sour-smelling mass to a solid bolus of mud, feces, and grain. Semiliquid contents have been most common. The contents usually suggest a depraved appetite.

**Autopsy findings.** Few or no changes in any organ except the crop and possibly the lower esophagus are seen on autopsy. The mucous membrane of the bulbous portion of the crop is thickened and in folds. Areas of diseased lung tissue varying in size are easily seen in those cases where the cause of death has been mechanical pneumonia caused by the entrance of crop contents into the lung. In such cases, food particles are found in the bronchi when the latter are carefully dissected. The air sacs are sometimes involved, and foreign matter can be seen, when scrapings from them are examined microscopically.

**Prevention, control, and treatment.** Since pendulous crops are associated with a hereditary weakness on the part of the individual, obviously the best preventive measure is to avoid mating any birds that have a family history of this weakness. Although this is a difficult procedure in the flock that is not trapnested, much can be done to prevent the condition from becoming established. Poults with affected crops should be caught and toe-marked or banded so that they can be eliminated at the time when turkeys are selected for breeding.

Sufficient shade during the hot months will reduce the numbers of pendulous crops in a flock. It is doubtful, however, whether any procedure other than eliminating the inherent tendency will remove the possibility of having a few cases.

Many methods for "curing" pendulous crops have been described by turkey growers. These have included various operations, the use of cloth vests or supporters, and methods of portioning out the water supply to the affected birds. Most of the methods that have come to the writer's attention, however, have produced few or no actual recoveries.

Removing a portion of the crop surgically results in a high percentage of recoveries, but the time consumed probably does not warrant the procedure as a routine practice. Washing out the crop (Fig. 40.51 A and B) with warm water containing a weak antiseptic, and then tying off a portion of the skin over the enlarged crop, also yields temporary relief until market time in a small percentage of cases (Fig. 40.52 A and B) . If only a few cases appear,

FIG. 40.51. A—the use of a veterinary stomach pump for douching a pendulous crop. B—draining the pendulous crop which had been filled by the method shown in A. (Hinshaw, Univ. of Calif.)
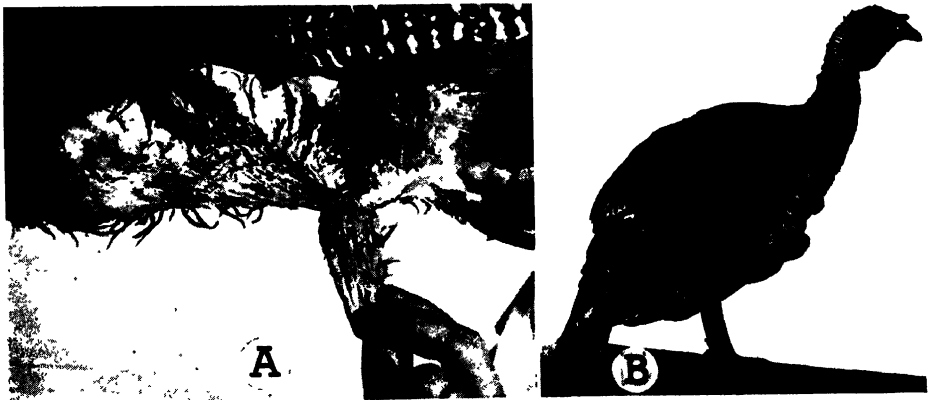


FIG. 40.52. A—method of tying off a section of skin to relieve pendulous crop. A suture is passed through the skin and then wound several times around before being passed through the skin a second time and tied. B—turkey three weeks after being treated as indicated in A. Note that the tied off section of skin has become necrotic and is about to drop off. (Hinshaw, Univ. of Calif.)

it is probably more economical to kill the affected birds than to attempt treatment.

## REFERENCES

Asmundson, V. S., and Hinshaw, W. R.: 1938. On the inheritance of pendulous crop in turkeys (*Meleagris gallopavo*). Poultry Sci. 17:276.

Hinshaw, W. R., and Asmundson, V. S.: 1936. Observations on pendulous crop in turkeys. Jour. Am. Vet. Med. Assn. 88:154.

## POISONING

Although losses from poisoning in turkey flocks are not great, a few cases are briefly described below. The tolerance of turkeys to rodent poisons is also discussed in answer to inquiries on this subject. In most outbreaks traced to poisoning, the symptoms and autopsy findings resemble those already described under the heading of enteritis. The diagnosis depends on discovering poison by chemical analysis of the crop or gizzard contents or on finding poison in the food supply.

**Arsenic.** DeLay (1940) reported losses in ten-week-old poults from eating grasshopper bait containing sodium arsenite and bran moistened with water. The bait was spread unevenly on a turkey range so that the birds had access to clumps of the mixture as well as to the grasshoppers. The owner reported a mortality of 5 per cent from the poisoning. DeLay fed some of the same mixture to eight-week-old poults in such a manner that the poults consumed from 0.25 to 0.5 gram of arsenic trioxide. Both dosages killed the experimental poults; the larger dose in 2 to 12 hours, and the smaller dose in 20 to 72 hours after being fed. The smaller dosage approximated that consumed on the ranch. The post-mortem findings in the field cases described by him were grasshoppers in the crop, hemorrhagic inflammation of the duodenum and jejunum, and a "sweetish" odor of the gizzard and intestinal contents. Arsenic was detected in the intestinal contents and in the grasshoppers found in the crop, by the Gutzeit method.

According to Whitehead (1934) arsenic in bran used for grasshopper poisoning is not present in sufficient amounts to produce mortality in birds, if the mixture is spread evenly and thinly over the ground. Growers are cautioned, however, to be sure that these recommendations are followed if such a procedure is necessary on a turkey range.

**Copper sulfate (bluestone).** According to experimental work by Hinshaw and Lloyd (1931), turkeys may be poisoned by copper sulfate added to the drinking water in concentrations greater than 0.2 per cent (1:500 dilution). As turkeys do not like copper sulfate solutions in any dilution and will avoid them if untreated water is present, poisoning is unlikely unless no other source of drinking water is available. In cool weather turkeys may go without drinking for several days rather than drink water containing even

nontoxic doses of this chemical. For these reasons, copper sulfate is not recommended except for specific uses, and in concentrations not exceeding 0.05 per cent (1:2,000 dilution). The poisoning is usually evidenced by a greenish-blue stain on the crop. Marked erosion of the mucous membranes follows excessive doses.

**Mercuric chloride (corrosive sublimate).** Mercuric chloride is well known for its toxic nature, but in spite of this is often carelessly used on turkey ranches as a disinfectant and remedy. It is too commonly recommended for treatment of drinking water without experimental basis for the recommendation.

A series of trials made on the toxicity of this chemical for turkeys by McNeil and Hinshaw (1945) showed that a 1:2,000 dilution as a sole source of drinking water was definitely toxic, killing five out of six eight-week-old poults in two weeks. One poult lived for two weeks under such treatment but was definitely undersized when killed. Eight-week-old poults tolerated 1:4,000 dilution for two weeks, but one poult out of ten died in a two-week-old group. Both groups given 1:4,000 dilution failed to make normal gains. Both two-week-old and eight-week-old poults tolerated 1:8,000 dilution for two weeks and made normal gains, but at necropsy the two-week-old group showed some necrosis of the gizzard lining.

The principal autopsy finding in mercuric chloride poisoning is a marked thickening and necrosis of the gizzard lining. There was some escharotic thickening in the crop, and the mucous membrane of the proventriculus was often sloughed. The poults given 1:8,000 dilution (which is the amount usually used by growers) for an extended period (two weeks) showed some thickening and necrosis of the gizzard membranes.

These results show the dangers encountered when using such toxic chemicals. Until more evidence is available on its value, mercuric chloride cannot be recommended for use on the turkey ranch.

**Poisonous weeds.** The fact that turkeys are often ranged among poisonous weeds suggests the reason for losses that are sometimes experienced on pasture lands. There are no experimental data available, however, on weed poisoning in turkeys. Where heavy losses occur in young turkeys reared on pasture, poisonous weeds should be sought as a possible cause. Suspected plants should be sent to a diagnostic laboratory together with tissue specimens for diagnosis and identification.

As a rule, animals or birds will not eat poisonous plants, unless other forms are not available. Most cases of poisoning result from the eating of young, growing shoots that come up in the spring before more palatable and nonpoisonous plants appear. Under certain conditions the seeds of poisonous plants may cause losses if accidentally mixed with grains.

The only method of control is to remove the cause. If the birds are rang-

ing in suspected areas, confining them in enclosures for a few days and supplying them with sufficient freshly cut greens is recommended. When they are again turned out on the range, the supply of fresh greens should be continued until the suspected poisonous plants have been replaced by nonpoisonous varieties.

Examples of poisonous weeds which have been known to cause losses in turkeys are the seeds of certain of the lupine, young shoots of oleander, and the second, succulent growth of Sudan.

Oleander (*Nerium oleander*) poisoning in turkeys is occasionally seen, but under normal feeding conditions poults will not eat even the young succulent shoots. In one experimental trial by McNeil and Denny (1939), five out of six four-week-old poults were killed by inserting leaves of oleander sprouts into their crops. The same poults had refused to eat the leaves when offered as greens. The poults died within 24 hours after the forced feeding, and at autopsy showed hemorrhagic enteritis. Three adult turkeys were fed young succulent oleander shoots for two weeks in lieu of greens. The birds continually refused them, even when they were cut up and mixed with the grain. The presence of oleander leaves in the crop and gizzard together with a history of the poults eating the plant is evidence of poisoning.

**Sodium bicarbonate (baking soda).** Sodium bicarbonate has been shown by several investigators (Delaplane, 1934; Hoffman, 1942; and Witter, 1936) to cause losses in chickens. These losses are manifested by lesions in the kidneys and other organs similar to those seen in gout. Hoffman found that the continuous use of amounts of sodium bicarbonate in excess of 15 grams per gallon of drinking water is toxic for baby chicks if used as a substitute for all other drinking water.

The toxicity of sodium bicarbonate for turkey poults from four to eight weeks of age has been determined at the California station (unpublished data). The results obtained were similar to those reported by the above investigators. When more than 0.6 per cent of sodium bicarbonate was given in the drinking water to four- and six-week-old poults, some mortality resulted, while eight-week-old poults were able to tolerate 1.2 per cent. Marked uremia and arthritis developed in all ages when over 0.6 per cent was given. As noted by Witter, sodium bicarbonate given in subtoxic doses also caused increased water consumption and diarrhea in turkey poults. Therefore, sodium bicarbonate is not a safe drug to use on the turkey ranch.

**Sodium chloride (common salt).** One outbreak of enteritis in turkeys about two-thirds grown finally proved to be associated with the use of well water containing a high percentage of common salt. This was the only source of water, and the losses probably resulted from heat prostration combined with salt dehydration; the turkeys did not like the water and drank only small quantities. A supply of fresh water stopped the losses within a few

days. Another instance of losses from enteritis probably due to salt consumption was traced to boxes of salt placed on the range for sheep that were being pastured with the turkeys (see also Ascites, page 1101).

**Strychnine.** Inquiries on possible poisoning by the strychnine-coated grain used for rodent control on cutover grain fields stimulated a series of experiments to determine the tolerance of turkeys for strychnine. Based on the results, turkeys will tolerate the usual amounts of strychnine in poisoned grain. Despite considerable variation in individual tolerance, there is probably little danger, provided other grain is available. Turkeys dislike grain coated with even minute amounts of strychnine and, after the first taste, will usually leave the planted poison bait alone and seek more palatable food.

**Miscellaneous.** Many other poisons could be mentioned, but they are not common causes of losses, and little is known about the exact tolerance of turkeys to them. Circumstantial evidence often points to poisoning when it is difficult to prove that a particular poison is responsible. Such chemicals as mercuric chloride, lead arsenate, and thallium, used occasionally on the farm, should be stored out of reach of turkeys. While chemical sprays or dusts are being applied in orchards where turkeys are ranging, the birds should be removed. After the orchard has been sprayed or dusted, there is still some danger from the residue on the covercrop; if other range is available, the birds should be kept out of the orchard for several additional days, or until a rain has reduced the residue remaining on the forage.

### REFERENCES

Delaplane, G. F.: 1934. Some of the tissue changes in poultry resulting from the ingestion of sodium bicarbonate. Vet. Alumni Quart. (Ohio State Univ.) 21:149.

DeLay, P. D.: 1940. Grasshopper-poison bait and turkey poult mortality. Jour. Am. Vet. Med. Assn. 97:149.

Hinshaw, W. R., and Lloyd, W. E.: 1931. Studies on the use of copper sulphate for turkeys. Poultry Sci. 10:392.

Hoffman, H. A.: 1942. Unpublished data, used by permission.

McNeil, E., and Denny, I.: 1939. Unpublished data, used by permission.

——— and Hinshaw, W. R.· 1945. Effect of mercuric chloride on turkeys and on *Hexamita meleagridis*. Poultry Sci. 24:516.

Whitehead, F. E.: 1934. The effect of arsenic, as used in poisoning grasshoppers, upon birds. Okla. Agr. Exper. Sta., Bul. 218.

Witter, J. F.: 1936. A preliminary report on the injurious effect of sodium bicarbonate in chicks. Poultry Sci. 15:256.

### NEMATODE, CESTODE, AND TREMATODE INFESTATIONS

Since most of the parasitic worms affecting turkeys are also common to chickens, the reader is referred to the general section for detailed descriptions of these parasites. Control and treatment are also discussed in the general section.

**Capillaria.** Of the several species of the genus Capillaria that infest domestic birds, at least three have been reported in turkeys. Two of these,

*C. annulata* and *C. contorta,* infest the upper digestive tract, while one, *C. columbae* (Graybill, 1924; Wehr, 1939a) is found in the intestines. Cram



FIG. 40.53. Typical penguin-like posture of a turkey in advanced stage of *C. contorta* infestation. (Emmel, Jour. A.V.M.A.)

(1926) reported *C. annulata* in turkeys in 1926 and later (Cram, 1936) published a comprehensive review on this and other species, giving the principal morphological characteristics of each. Emmel (1939) has described symptoms and autopsy findings observed in three outbreaks due to *C. contorta.* He calls attention to the penguin like attitude of infested turkeys (Fig. 40.53). Figure 40.54 shows the gross lesions observed in one of Emmel's specimens. He reported that 5 per cent commercial flowers of sulfur fed in the regular mash caused marked improvement of infested birds in 4 days and prevented new cases from appearing in the flock. The injurious effect of prolonged administration of sulfur is discussed in the sections dealing with coccidiosis and nutrition.

**Cecal worms.** Cecal worms (*Heterakis gallinae*) are of importance because they act as carriers of *Histomonas meleagridis,* the causative organism of blackhead. Recommendations for their prevention will be found under Blackhead.

**Gapeworms.** Gapeworms (*Syngamus trachea*) cause some mortality in young turkeys, and as shown by Wehr



FIG. 40.54. Crop and esophagus of a turkey suffering from *C. contorta* infestation. (Emmel, Jour. A.V.M.A.)

(1939b), survivors may carry the parasite for as long as 224 days. Such survivors are important means of transmission to susceptible chickens and poults.

**Ascarids.** The intestinal roundworm, *Ascaridia galli,* is not an important parasite of turkeys, and under good husbandry practices the turkey grower need not fear losses from it. Examination of diagnostic records from avian pathology laboratories shows that the incidence of even mild infestations is less than 1 per cent of all specimens examined. Evidence that the turkey is more resistant to the *Ascaridia galli* than is the chicken has been reported by Ackert and Eisenbrandt (1933). Remedies for control of these parasites are not recommended unless definite evidence of a severe flock infestation is found.

**Tapeworms** may present an economic problem to the turkey grower. Most
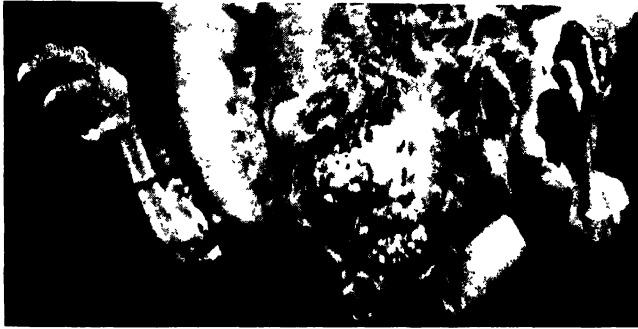


Fig. 40.55. A six-week-old turkey poult showing perianal and abdominal groups of cysts of a fluke *Collyriclum faba.* (Riley and Kernkamp, Jour. A.V.M.A.)

of the cestodes that infest chickens are also parasitic for turkeys. Prevention, control, and treatment are similar to those outlined for chickens.

Combination anthelmintics designed for removal of both roundworms and tapeworms are not recommended for turkeys. Under no condition should treatment for either type be instituted unless the parasites are known to be causing losses in the flock. Emphasis should be placed on prevention, not on treatment.

**Flukes.** Riley and Kernkamp (1924), Riley (1931), and Marotel (1926) reported a monostome fluke, *Collyriclum faba,* which encysts in the skin of turkeys and other birds. These usually are found in the abdominal region, and especially in the perianal region, with occasional cysts on other parts of the body (Fig. 40.55). This fluke has been reported in many species of birds by other investigators. Although the complete life history has not been determined, Riley (1931) believes that snails probably act as the first intermediate host, and nymphs of dragon flies as the second intermediate host. English sparrows appear to be important disseminators of the parasites.

Annereaux (1940) reported the occurrence of typhlitis in poults caused by a fluke, *Echinoparyphium recurvatum* (von Linstow). The ten-week-old poults involved in this outbreak were being ranged along a creek where two

types of snails and many tadpoles were present. The lesions found in the ceca of the affected poults were characteristic of those seen in the ceca of poults suffering from blackhead, but no liver lesions were noted. Foggie (1937) has reported an outbreak of parasitic necrosis of intestines of turkey poults in Ireland caused by a fluke, *Plagiorchis laricola* (Skrjabin), normally a parasite of terns and gulls.

According to Macy (1939) the most important species of trematode parasites for North American poultry is *Prosthogonimus macrorchis*. Although not found in natural outbreaks, Macy was able to infect turkeys with this parasite; typical lesions were observed in the oviducts of the parasitized birds. Few external symptoms of disease were noted, but the turkeys ceased laying 4 days after being fed the parasites. This trematode is transmitted by dragonfly nymphs (Lakela, 1932).

No satisfactory treatment has been reported for trematodes in turkeys. Prevention of infestation consists in avoiding access to marshy pastures, lake shores, or infested streams.

## REFERENCES

Ackert, J. E., and Eisenbrandt, L. L.: 1933. On the comparative resistance of Bronze turkeys and White Leghorn chickens to the nematode *Ascaridia lineata* (Schneider). Jour. Parasit. 20:129.

Annereaux, R. F.: 1940. A note on *Echinoparyphium recurvatum* (von Linstow) parasitic in California turkeys. Jour. Am. Vet. Med. Assn. 96:62.

Cram, E. B.: 1926. A parasitic disease of the esophagus of turkeys. No. Am. Vet. 7:46.

———: 1936. Species of Capillaria parasitic in the upper digestive tract of birds. U.S.D.A., Tech. Bul. 516.

Emmel, M. W.: 1939. Observations on *Capillaria contorta* in turkeys. Jour. Am. Vet. Med. Assn. 94:612.

Foggie, A.: 1937. An outbreak of parasitic necrosis in turkeys caused by *Plagiorchis laricola* (Skrjabin). Jour. Helminth. 15:35.

Graybill, H. W.: 1924. *Capillaria columbae* (Rud.) from the chicken and turkey. Jour. Parasit. 10:205.

Lakela, O.: 1932. Chickens definite hosts to species of Prosthogonimus. Poultry Sci. 11:181.

Macy, R. W.: 1939. Disease in turkeys due to *Prosthogonimus macrorchis*. Jour. Am. Vet. Med. Assn. 94:537.

Marotel, G.: 1926. Une nouvelle maladie parasitaire: La monostomidose cutanée du dindon. Rev. vét. 78 (12) :725.

Riley, W.A.: 1931. *Collyriclum faba* as a parasite of poultry. Poultry Sci. 10:204.

——— and Kernkamp, H. C. H.: 1924. Flukes of genus Collyriclum as parasites of turkeys and chickens. Jour. Am. Vet. Med. Assn. 64:591.

Wehr, E. E.: 1939a. Studies on the development of the pigeon capillarid, *Capillaria columbae*. U.S.D.A., Tech. Bul. 679.

———: 1939b. The gapeworm as a menace to poultry production. Proc. Seventh World's Poultry Cong. P. 267.

## ECTOPARASITES

Reference is made to the general section on External Parasites for a detailed discussion, including prevention and control.

**Lice.** Turkeys may be infested with the common body louse of chickens, *Eomenacanthus stramineum* (*Menopon biseriatum*) and the chicken shaft

louse, *Menopon gallinae.* The large turkey louse, *Goniodes meleagridis,* and the slender louse, *Lipeurus gallipavonis,* are probably native to the turkey. The large turkey louse is the most common. Rearing turkeys in close confinement, and insanitary quarters, favor lice more than does range rearing. It is important that breeding males and females be examined frequently for lice, since parasites may be a very important cause of infertility. A common method of introducing lice to an uninfested ranch is by the use of infested shipping crates that have been brought on the ranch by buyers. Growers should insist that buyers clean and disinfect all crates and equipment used to transfer stock to killing plants.

**Mites.** Turkeys are less affected with mites than chickens, due probably to the greater tendency to rear turkeys out of doors. Chickens reared in close proximity to turkeys, and the use of old chicken yards constitute the chief sources of infestation.

**Ticks.** The only tick of economic importance is the fowl tick or "blue bug," *Argas persicus.* Spirochaetosis, a disease transmitted by this tick, has recently been diagnosed in the United States by Hoffman, Jackson, and Rucker (1946) in turkeys, and by Burroughs (1947), in chickens. For more details, see the section on spirochaetosis in turkeys, page 1070. It is important that all fowl to be purchased, be inspected for the presence of these parasites. Birds (chickens and other fowl as well as turkeys) from infested ranches should never be brought on tick-free ranches.

### REFERENCES

Burroughs, A. L.: 1947. Fowl spirochetosis transmitted by *Argas persicus* (Oken), 1818, from Texas. Science 105:577.

Hoffman, H. A., Jackson, T. W., and Rucker, J. C.: 1946. Spirochetosis in turkeys (a preliminary report). Jour. Am. Vet. Med. Assn. 108:329.

# INDEX